THE INTRODUCTION OF NATIVE FOREST FLOOR PLANT SPECIES INTO THE INDUSTRIALLY DISTURBED FORESTS OF SUDBURY, ONTARIO, CANADA

by

Kierann R. Santala

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APPROVED/APPROUVÉ

Thesis Examiners/Examinateurs de thèse:

Dr. Peter Ryser
(Supervisor/Directeur de thèse)

Ms. Lisa Léger
(Committee member/Membre du comité)

Dr. Stephen Monet
(Committee member/Membre du comité)

Dr. Peter Beckett
(Committee member/Membre du comité)

Dr. Daniel Campbell
(Committee member/Membre du comité)

Dr. Gerard Courtin
(Committee member/Membre du comité)

Dr. M. Anne Naeth
(External Examiner/Examinatrice externe)

Approved for the School of Graduate Studies
Approuvé pour l'École des études supérieures
Dr. David Lesbarrères
M. David Lesbarrères
Director, School of Graduate Studies
Directeur, École des études supérieures

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Abstract

This study investigated the transplantation of understory plants within the Cu-Ni smelter-damaged urban forest of Sudbury, Ontario, Canada, to increase plant biodiversity in an area where natural colonization of understory species is delayed. The goal of my study was to evaluate establishment of 16 m^2 vegetation mats along a gradient of smelter disturbance and to relate successful establishment to abiotic and biotic site characteristics. Specific investigations were conducted to determine whether soil quality influenced root growth and transplant establishment. Variables associated with smelter emissions and soil temperature were the best predictors of successful transplant establishment of understory plant species, but relationships were species specific. Also, root growth was not limited to organic soils of the transplant mat and roots were able to grow into receptor site soil. Knowledge of environmental factors influencing establishment will help to determine site locations and to select species to introduce when transplanting understory species in future reclamation projects.

Keywords

Transplant success; reintroduction; understory plants; temperate and boreal forest; environmental site characteristics; ecological restoration; mine site restoration.

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Chapter 1

1. Introduction

The leading threat to the world's biodiversity is habitat loss and fragmentation of natural systems by humans for land use, such as mining or forestry (Tabarelli & Gascon 2005). Biodiversity can be affected throughout the lifecycle of a mine by the clearing of land, and through dust and smelter emissions (International Council on Mining and Metals 2006). The forest industry can also have lasting effects on biodiversity with some species that should typically be found in naturally regenerated forests remaining absent in 50 year old plantations (Aubin *et al.* 2008). With the decline of natural, unmanaged forests there is an interest in developing management strategies to improve the species composition, structure and function of human-created forests with the aim that one day they will support desired biodiversity (Aubin *et al.* 2008; Brudvig 2011).

The area of focus in this investigation, the copper-nickel smelter impacted urban forests of Sudbury, Ontario, Canada, is an example of the negative effect that past logging and copper-nickel smelting activities and fire can have on ecological diversity (Winterhalder 1995). Studies within this region have found that plant diversity is reduced in metal-contaminated areas surrounding smelters as a result of emissions (Amiro & Courtin 1981). The legislation for land restoration of many provinces and territories recommend that re-vegetation promotes the establishment of self-sustaining vegetation that resembles that of the natural environment, with the exception of erosion prone areas or areas with alternate land management objectives (Native Plant Working Group 2000; Manitoba Industry Trade and Mines 2006; Ministère des Ressources naturelles du Québec and the Ministère de l'Environnement et de la Faune du Québec 1997;

Ministry of Energy; Mines and Petroleum Resources Mining and Mineral Division 2008; Indian and Northern Affairs Canada 2007; Ontario Mining Act 2009; New Brunswick Department of Natural Resources 2012). Traditionally, re-vegetation of mine lands consists of planting trees and letting understory species recolonize these areas naturally (Mackenzie & Naeth 2010). However, succession of understory species has been found to be delayed in restored forests due to poor dispersal ability of these species (Braun & O'Hara 2006 *unpublished data*; Mackenzie & Naeth 2010). In the Sudbury region, over 30 years of reclamation activities have allowed young forest to develop, yet understory diversity in this region remains low (Braun & O'Hara 2006 *unpublished data*; City of Greater Sudbury 2012b; Monet *et al.* 2012). The goal of my investigation is to identify site characteristics associated with successful establishment of forest floor species introduced in the form of transplants. This information will be useful for site selection when transplanting understory species in the future, potentially leading to improved regreening efforts in both boreal and temperate forests.

Forest floor plant species (or understory species) in my investigation include plant species occupying the lower-most strata of the forest and includes herbs, shrubs, lichens, mosses and tree seedlings. These species play important functional roles in forest ecosystems including the maintenance of water quality (Naiman & Décamps 1997), soil stability (Pierson *et al.* 2007) and forest nutrient status (Muller 2003), and the recovery of forests following disturbances (Muller 2003; Isbell *et al.* 2011). Species-specific examples of functional roles of understory plants are discussed in the following chapter.

The transplantation of native plant species is becoming a popular method to overcome barriers associated with dispersal (Sheddon 2010); however, many native plant species are not commercially available. Those species that are available might be cost-prohibitive to use on a landscape scale. There are also some other challenges to their use, for example their poorly known ecological requirements. Only a few plant reintroduction studies have been published in the scientific literature (Bottin *et al.* 2007). Consequently, information regarding the establishment of understory species is rare. Even more rare are studies exploring the reintroduction of understory species on metal contaminated soils. Braun (2007) and Winterhalder (2004) compared methods used to reintroduce temperate and boreal species on smelter-impacted forests and on reclaimed mine tailings, whereas Koch (2007) reviewed techniques used to restore understory vegetation in a Bauxite Mine in Western Australia. Because Canada has a large mineral processing industry with 30 non-ferrous metal smelters and refiners (Marshall 2012), there is an interest in testing the application of temperate and boreal understory transplants in metal contaminated soils. In the following chapter of this thesis I will discuss ethical considerations related to the transplantation of native plant material for reclamation purposes. I will also review techniques associated with successful transplant projects.

Despite refinements in reintroduction techniques, gaps still remain in our knowledge of native plant establishment, particularly in regards to determining their habitat requirements. A major source for transplant failure is the placement of target species into areas that do not fulfill their ecological or biological requirements (Godefroid *et al.* 2010; Drayton & Primack 2012). Consequently, there is a need to determine the biotic and abiotic site requirements for species used in restoration (Drayton & Primack 2012). A review of literature regarding the general autecology of understory species is provided in the next chapter. Although this literature provides important information regarding how biotic and abiotic forest characteristics can influence understory biodiversity, one should be cautious about generalizations in regard to the effects of site characteristics on understory diversity because of differences in species composition, land management, and site attributes (Barbier *et al.* 2008).

I asked two main questions in this investigation (i) *What are the important environmental site characteristics associated with the successful establishment of understory species transplanted along a gradient of metal contamination?* (ii) *Would newly transplanted species be capable of growing roots in soils with elevated metals?* To answer these questions, I monitored 30 forest floor transplant plots (16 m^2 in size) located in a range of metal contaminated soils and forest types in the smelter disturbed forests of Sudbury, Ontario, Canada. Environmental site characteristics in this investigation consisted of plant-available chemicals of concern, plant-available nutrients, canopy cover and soil temperature. Establishment was evaluated by comparing measures of community composition and the spread of species from the plot. As well, for three select species – Canada mayflower (*Maianthemum canadense*)*,* wintergreen (*Gaultheria procumbens*) and blue-bead lily (*Clintonia borealis*) – root growth and sexual reproduction within plots were also observed. I also compared root growth of wintergreen (*Gaultheria procumbens*) and blue-bead lily (*Clintonia borealis*) grown in donor and receptor site soils in plots located within two smelter disturbance zones, barren (highly affected by the smelter) or semi-barren (less affected by the smelters).

Literature regarding the importance of environmental requirements of transplanted forest floor species is inconsistent. For example, Mottl *et al.* (2006) suggested that detailed knowledge of the variation in light availability and other abiotic factors might not be needed for successful restoration of woodland herbs. In contrast, Peterson & Philipp (2001) who observed the establishment of 37 herbaceous forest plants transplanted into stands of *Fagus sylvatica L.* in Denmark found that soil, light and moisture conditions at receptor sites were important for transplant establishment. A review by Trusty & Ober (2011) also found that survival of transplanted understory species on the coastal plains of the Southern United States was greater in forests with un-thinned canopies due to these species' adaptation to shade. Mackenzie & Naeth (2010) also found that total vegetation cover of transplanted upland boreal understory species was best correlated with soil organic matter content for a project occurring in Alberta, Canada. In the second chapter of this thesis, I will review of the autecology of my three select species, which will provide some indication as to the habitat preferences of these plants. However, these studies took place mainly in the Northeastern United States and Southeastern Canada; hence, the climatic conditions and geological history of these sites are different from those characterizing my sites.

Based on these studies I hypothesized that predictors related to light, microclimate and soil characteristics would be able to explain variation in transplant success. I predicted that root growth and sexual reproduction of observed species would be positively related to characteristics typical to that of an undisturbed forest with sufficient canopy cover, organic matter content and soil nutrient status. I also hypothesized that plants in plots located closer to the smelter would be limited in their performance due to low organic matter, low pH, and higher metal content at these sites. However, I expected that some understory species would be more tolerant to metal stress and would be capable of occupying these areas without being limited in their performance. For example, I predicted stress tolerant species such as wintergreen (*Gaultheria procumbens*) would produce more roots than blue-bead lily (*Clintonia borealis*). I also predicted that plant community characteristics such as Shannon-Wiener Diversity Index, evenness and species richness of the plots would also be highest on plots farthest away from the smelters.

This study will provide insight into the restoration of understory biodiversity. Information gathered will aid in site selection when introducing understory plant species in reclaimed forests of smelter-disturbed lands. Because the colonization of understory species in reclaimed forests of the Sudbury region is constrained due to the fragmented nature of this city, the assisted migration of these species through transplantation will give an idea as to the extent to which understory species are capable of colonizing metal-contaminated areas. It will also assist in the conservation of species by identifying ways in which human-made ecosystems can be managed to promote the establishment of understory species.

Chapter 2

2 Literature Review

The transplantation of native plant species is becoming a popular method to overcome barriers associated with dispersal (Sheddon 2010); however, only a few plant reintroduction studies have been published in scientific literature (Bottin *et al.*, 2007). In the following sections I will discuss topics relating to the use of understory plant species in land reclamation and how they will be applied to my study. These topics include:

1) The importance of understory species in the forest ecosystem
2) Ethical and technical considerations when introducing native understory species
3) Current research needs required to improve understory reintroduction
4) The influence of environmental site characteristics on understory species in general
5) Autecology of wintergreen (*Gaultheria procumbens*), blue-bead lily (*Clintonia borealis*) and Canada mayflower (*Maianthemum canadense*)

2.1 The importance of understory biodiversity in the forest ecosystem

Our view of forests is often restricted to the trees that make up the canopy. However, the majority of plant biodiversity is actually in the understory layer that makes up more than 80% of plant species richness of the forests (Gilliam 2007). Literature has shown that plant species occupying this layer provide important ecosystem services in forests that include maintaining water quality (Naiman & Décamps 1997), soil stability (Pierson *et al.* 2007), forest nutrient status (Muller 2003), and the recovery of forests following disturbances (Muller 2003; Isbell *et al.* 2011).

Understory plants play an important role in maintaining water quality in riparian environments by buffering the effects of man-made pollutants. The Environmental Protection Agency (2012) has identified non-point source pollution as the leading cause of water quality problems. This is pollution that typically enters rivers and lakes when rainfall, snowmelt or irrigation runs over land or through the ground and picks up pollutants. Eventually these pollutants are carried to rivers and lakes where they become deposited (Kloiber 2006). Riparian forests and streamside grasses effectively prevent sediments and sediment-bound pollutants from entering stream water by trapping sediment (Naiman & Décamps 1997). Runoff after low intensity rainfall events has been found to lead to rapid soil erosion in areas with low understory cover. Enhancing understory cover can prevent soil erosion by increasing storm water infiltration capacity and reducing the velocity of overland flow (Pierson *et al.* 2007). For these reasons, the supplemental planting of vegetation to restore the stabilization of contaminated soils is an effective means of reducing pollution from entering water bodies (Wong 2003).

Understory plants also play an integral role in the cycling of forest nutrients. This is the result of their high foliar concentration of nutrients, the rapid rate at which their litter decomposes and high turnover rates of their short-lived above ground biomass (Muller 2003). Moreover, this vegetation can act as a potential sink for vernal nutrients in northern hardwood forests (Tessier & Raynal 2003). For example the spring ephemeral trout lily (*Erythronium americanum* Ker Gawl.), is known to reduce the loss of potassium and nitrogen by taking up these nutrients prior to the deciduous tree canopy developing; a time when nutrient loss is the greatest due to snowmelt runoff and low nutrient uptake by deciduous species (Muller & Bormann 1976; Knight *et al.* 1985).

Understory plants also play a role in the resilience of forests to disturbances such as fire, insect attacks and climate change. Studies have found that following oak (*Quercus sp.*) defoliation or mortality caused by gypsy moth (*Lymantria dispar* L.) within oak dominated forests, the composition of trees making up the canopy shifts and becomes more diverse. Understory trees such as red maple and sugar maple expand their crowns due to the increase in light availability caused by new overstory canopy gaps. The expanded crowns of understory trees then cause intense light competition for regenerating oak seedlings resulting in their mortality. As a result, following insect attacks oak dominated forests recover through the development of tree species that make up its understory (Collins 1961; Fajvan & Wood 1996). Plant species-richness, defined as the number of different species that can be found in a particular area (Adams 2009), is also potentially beneficial in a changing climate. If the climate changes and gives rise to adverse conditions for a given species that performs an important role, ecosystem functions can be maintained by another functionally similar species better adapted to the new environmental conditions (Isbell *et al.* 2011).

Diverse understory plant communities are important in forests because these plants maintain water quality, soil stability, nutrient status and aid in the resilience of forest following disturbances (Naiman & Décamps 1997; Muller 2003; Pierson *et al.* 2007; Isbell *et al.* 2011). As land restoration seeks to return degraded lands to sustainable and functional ecosystems (Bradshaw 1984) there is an interest in the reintroduction of understory species that can perform these functions in reclaimed forests.

2.2 Ethical and technical considerations when introducing native understory species

Because plant reintroductions are costly, labour-intensive projects that are not always successful (Godefroid & Vanderborght 2011) there are times when the reintroduction of native vegetation is not recommended. For example, non-natives are advantageous in badly degraded sites where it is difficult or impossible to establish native species (Lamb 1998). Due to poorly understood ecological requirements of native plants, particularly rare ones, there are also concerns over their relocation (Fahselt 2007). The transplantation of rare or isolated populations is not recommended by the Society of Ecological Restoration due to risk of inbreeding (SER 2010). There are also concerns that once transplantation is proposed, habitat destruction becomes more acceptable (Fahselt 2007). Because current restoration techniques are unable to recreate ecosystems, destruction of habitat cannot be justified with the use of restoration ecology (Brudvig 2011). The Ontario Chapter for the Society of Ecological Restoration's guidelines for the use of indigenous species in restoration projects state that nursery propagated plant material be used whenever possible and if not available that seed and cuttings be used in preference to the wild collection of the entire plant. The salvaging of plants from an area to be destroyed is only permitted when there is no possibility that a development plan will change and only species that are known to survive transplanting should be collected (SER 2010).

The age at which transplanted species are relocated has a profound influence on the success of establishment (Godefroid *et al.* 2011). Studies have found greater success rates of mature plants than those of seedlings or seeds (Godefroid *et al.* 2011; Drayton & Primack 2012). Older plants have been found to have higher survival rates and grow larger than young seedlings because young plants are more susceptible to harsh environmental conditions due to their small size (Kindell *et al.* 1996). Transplanting mature plants also bypasses the hazards associated with germination of seeds in the field (Davey 2002). There are some exceptions to these findings. For example, introductions of big sagebrush plants (*Artemisia tridentala* Nutt.) and lodgepole pine (*Pinus contorta* Douglas ex Loudon var.*latifolia* Engelm) introduced using seeds were found to produce more seeds and had more above-ground growth than those planted as seedlings (Welch 1997; Robert & Lindgren 2006). Although the planting of mature vegetation appears to be the most successful reintroduction method based on reviews of literature (Godefroid *et al.* 2011), the best method of establishment is likely species specific.

There is evidence that using mats of vegetation that contain both multiple plant species and underlying soil is the most effective means of reintroduction. Braun (2007) found that the survival and the number of established understory species was greater for plants that were introduced into smelter disturbed forests when reintroduced using mats of vegetation and their associated soil (also referred to as sods) compared to introductions by importing topsoil or transplanting plugs of single individuals. Similar findings have been found for prairie (Kerns 1986) and heathland restoration projects (Pywell *et al.* 1995). Other studies suggest that the best method of introduction might be species-specific. Winterhalder (2004) found in an 8-year study that some understory plants performed better when introduced in the form of mats of vegetation (sods) whereas other species established better when introduced in topsoil heaps. For example, Canada mayflower (*Maianthemum canadense* Desf.) and barren ground strawberry (*Waldstenia fraganoides* (Michx.) Tratt.) spread better when introduced in the form of plugs. In contrast, spreading dogbane (*Apocynum androsaemifolium* L.) and sweet fern (*Comptonia peregrina* (L.) J.M. Coult) were more successful when introduced in the form of topsoil containing plant propagules. It must also be kept in mind that introducing species using mats of vegetation is the most costly method and causes the greatest damage per area of the site in which donor material is collected.

Date of planting also plays a role in transplant establishment. A review of 17 forest groundcover reintroductions in the South Eastern United States found that the greatest survival rates corresponded to planting dates that occurred during seasons with low water and heat stress for newly planted species (Trusty & Ober 2011). Braun (2007) had similar findings in which forest floor mats planted in metal-contaminated soils in the fall contained more species in their third growing season than those planted in the spring. This finding was attributed to fall-planted forest floor mats having more time to establish prior to summer drought. A study from Wisconsin found that weather conditions at the time of planting had more of an effect on transplant survival of prairie vegetation than the time of year (Kerns 1986). This study recommended that transplanting could occur at any time of the growing season provided it was done during cool, cloudy and humid conditions.

Studies regarding thickness of soil application for transplant establishment are limited to studies using topsoil as a propagule source. Determining the thickness of soil application appears to be a balance of enhancing conditions for seed germination while at the same time promoting favourable growing conditions for seedlings. Koch (2007) recommended for restoring understory vegetation native to the Jarrah forest of Australia that topsoil be applied at a thickness no greater than 15 cm to avoid seeds from becoming buried too deep to germinate. To restore heathland species it is recommended that topsoil be collected at a depth of 5 cm or less and spread at a depth of 2.5 cm to allow for adequate regeneration (Putwain & Rae 1988).

In contrast, thick soil applications may enhance growth once reintroduced individuals become established. A study by Mackenzie & Naeth (2010) which reintroduced understory boreal plants on reclaimed mine lands found that a thick topsoil treatment of 20 cm promoted favourable growing conditions that allowed for greater cover of introduced species compared to that of the thinner treatments at 10 cm. Higher densities of upland boreal plants in the 20 cm thickness treatment was attributed to less mixing with underlying subsoil during application and fewer bare areas created during the application process. Reduced seed germination in 10 cm treatments was thought to be caused by greater amounts of mineral soil on the surface due to mixing of this treatment which would have decreased light penetration and lowered nutrient availability (Mackenzie & Naeth 2010). These studies suggest that when determining thickness of soil application, one should have soils thin enough to promote seed germination but not so thin as to reduce the conditions for favourable growth once seedlings become established. Thickness of soil will also depend on the project objective, the amount of donor material available, area to be restored, and rooting depth of species used. Hence, thickness of soil application used should be tested to determine a site-specific method.

To enhance establishment rates, having a well thought-out site maintenance strategy before and after transplantation is recommended by the literature. Survival rates were found to be significantly greater for reintroduced species placed into fenced areas rather than in unprotected areas as it prevented them from being consumed by herbivores (Godefroid *et al.* 2011). Thinning of plants surrounding the plots can be an effective means of reducing plant competition, but can also lead to alterations in light condition. For example, forest floor vegetation transplanted into forests with un-thinned canopies had higher survival rates than those planted into areas with thinned canopies because of the adaptation of these species to shade (Trusty & Ober 2011).

Studies regarding the use of herbicides to prepare sites for plantings are not conclusive. Herbicide application has been found to decrease the survival of transplanted individuals, but increase the establishment of native seeded plants. These findings were related more to site characteristics than application of herbicide itself. Further research is needed to determine the number, timing and strength of application of herbicide to understand its true effects for site preparation for native plant establishment (Trusty & Ober 2011). Project developers who include site management strategies into their project design may reflect a greater commitment to the reintroduction project. This high level of commitment may also contribute to the greater success rates in projects that include a site management strategy (Godefroid *et al*. 2011).

2.3 Current research needs required for refinement of understory reintroduction techniques

A major source for transplant failure is the placement of target species into areas that do not fulfill their ecological or biological requirements (Godefroid *et al.* 2011; Drayton & Primack 2012). Not much information is available in the literature regarding the habitat requirements of native species (Mottl *et al.* 2006). As a result there is a need to determine the biotic and abiotic site requirements (Drayton & Primack 2012). This knowledge helps when selecting site locations for native transplantations for restoration purposes by providing environmental characteristics positively correlated with transplant establishment.

2.4 Influence of environmental site characteristics on understory species in general

Many studies have identified biotic and abiotic environmental characteristics associated with understory diversity (Huebner *et al.* 1995; Hart & Chen 2006) and have examined how to enhance this biodiversity through changes to the way forest plantations are managed (Newmaster *et al.* 2006; Macdonald & Fenniak 2007; Duguid & Ashton 2013). Light is considered a limiting resource when it comes to understory plant establishment and growth. Availability of this resource is controlled by the composition and structure of overstory trees and shrubs (Chen 1997; Meekins & McCarthy 2000; Yu & Sun 2013). It is widely accepted that forests with more tree canopy species also tend to have a more diverse understory (Gilliam 2007). Litter quality also affects site characteristics, such as soil nutrients and pH, and therefore plays a role in understory composition (Hart & Chen 2006). Studies in Northwestern Quebec found that nutrient availability under aspen, birch, spruce-fir and pine canopies differed due to the effect of canopy type on humus, pH, ion exchangeability and available calcium, magnesium, nitrate and phosphorus at these sites (Légaré *et al.* 2001). In general, deciduous litter has higher base cation contents and pH and decomposes more rapidly than coniferous species, and creates favourable conditions for vascular plant growth (Hart & Chen 2006). However, soil quality alone can also affect species composition. Légaré *et al.* (2001) found that soil texture influenced understory richness, evenness and composition, but not diversity. Species such as *Mitella nuda*, *Rubus pubescens* and *Galium triflorum* were associated with clay deposits whereas *Diervilla lonicera* and *Maianthemum canadense* were associated with till deposits. Clay deposits had higher species richness but diversity did not increase, suggesting that understory on clay soil had some species that were more dominant than that on till soils. Lamarche *et al*. (2004) found till in the area of the Légaré *et al.* 2001 study to have lower pH, base cation saturation, and C mineralization. Topography also affects diversity, with diversity being lower on south-facing slopes due to drought stress caused by higher evaporation rates at these sites (Huebner *et al.* 1995; Hart & Chen 2006). Generalizations about the effects of site characteristics on understory diversity is cautioned as studies have varying species composition, land management and site attributes (Barbier *et al.* 2008).

2.5 Autecology of wintergreen (*Gaultheria procumbens*), blue-bead lily (*Clintonia borealis*) and Canada mayflower (*Maianthemum canadense*)

The following section will provide information regarding the autecology of three species that were specifically observed for growth characteristics during this experiment. The reason that these three species were observed was because they were common in experimental plots and are known to occur within the Sudbury region.

***Gaultheria procumbens* L.** is a sun-tolerant evergreen shrub of the family Ericaceae that is common in closed-canopy coniferous woodlands, mixed wood and hardwood forests (Donohue *et al.* 2000; Roberts & Lixiang 2002; Moola &Vasseur 2009; Trock 2009). It is considered to be self-fertilizing, but is also a facultative out-crosser pollinated by insects, the majority of which are bumblebees (Mirick & Quinn1981). Rooting depth is limited to the litter layer and this species can spread vegetatively through rhizomes between 10 and 43 cm per year (Sobey & Barkhouse 1977; Flinn & Pringle 1983; Donohue *et al.* 2000)

Very little information exists on the habitat requirements of *Gaultheria procumbens.*

Only one study looked at the recovery of this species in logged forests of Eastern Canada. As mentioned previously this species can be found in a wide range of forest types. Studies have found this species to change growth patterns in response to canopy cover. In logged habitats that are resource-rich and open, *G. procumbens* grows in the form of coalesced clumps whereas in shaded habitats of late successional forests it grows in long chains. The compact growth form is thought to exploit favourable patches. This species also has an affinity for acidic soil, and grows well on many soil types of low nutrient status, including peat, sand, and sandy loam (Moola & Vasseur 2009). According to Rayfield *et al.* (2005), this species is associated with undisturbed sites in the Sudbury region (15 to 35 km from the smelters). However, based on vegetation surveys performed within the region it can be found within the Sudbury area at distances as close as 5 km from the Copper Cliff smelter and it appears to grow more densely in more open areas, for example in typically open canopy areas along rights-of-way (Santala & Popp 2010, *unpublished data*).

Clintonia borealis **(Aiton) Raf.** is a clonal, perennial forest herb that is restricted in range to the northern hardwood forests of Eastern North America (Angevine & Handel 1986). It grows in association with arbuscular mycorrhizal fungi of the family Glomeraceae (DeBellis & Widden 2006). *C. borealis* leaves develop in early spring and expand fully in June. It is capable of both self-pollination and cross-pollination; however, seedlings of this species are rarely observed (Angevine & Handel 1986). Typically this colonial plant reproduces vegetatively by ramets that are linked together by rhizomes that can draw upon stored reserves in times of growth, or supply the needs of other ramets in times of stress. Growth of this species is fast in the first year (6 cm $year^{-1}$) but slows in successive years (2 cm $year^{-1}$) (Angevine & Handel 1986; Dorken & Husband 1999). Rooting depth of this species ranges between 4 and 8 cm (Mallik &

Karim 2008).

Ashmun & Pitelka (1985) have shown through transplant experiments that took place in North-Eastern United States that *C. borealis* can survive in a wide variety of environments. It was found that genets collected from two shaded areas were larger during the first two years of growth when transplanted into a shaded garden in comparison to those placed in well-illuminated gardens. However, once established, growth was comparable between well-illuminated and shaded gardens. Initial reduced growth in open areas was thought to be caused by greater transplant shock caused by desiccation. This contradicts a study by Hughes & Fahey (1991), which also took place in the North Eastern United States, that found the frequency of *Clintonia borealis* decreased following the removal of shrub overstory. It has been suggested that natural populations do not tend to occupy well-illuminated areas because they are outcompeted by species better adapted to high light environments (Ashmun & Pitelka 1985). A study that investigated morphological differences in *Clintonia* sp. along an altitude gradient in Northern Vermont found that plants were the smallest at the transitional zone between the coniferous species of the upper slope and the deciduous canopy below. Environmental characteristics differed with elevation; for example, the upper slope had lower temperatures, greater precipitation and more acidic soils due to conifer cover, but this study was unable to show whether biotic or abiotic processes were the more important factors explaining growth (Carter & Vogelmann 1968).

Based on vegetation surveys within the region *Clintonia borealis* species can be found in the Sudbury region, but it tends to be infrequent in areas more strongly affected by smelter emissions and usually occurs in restored areas farther away from the smelter (approximately 5 km) (Santala & Popp 2010, *unpublished data*).

Maianthemum canadense **Desf.** is a perennial forest-understory herb that can be found in a variety of forest types (Silva *et al.* 1982). In central New Brunswick, flowers bloom in late spring and fruits appear in late summer (Helenurm & Barrett 1987). It is self-incompatible and it requires insects such as hoverflies and bees for pollination (Worthen & Stiles 1986; Hisatomo *et al.* 2008). *M. canadense* can spread quite quickly in comparison to other herbs spreading through rhizomes between 15 to 30 cm per year (Sobey & Barkhouse1977; Silva *et al.*1982). Rooting depth of this species is limited to the litter layer (Flinn & Pringle 1983).

Although *Maianthemum canadense* can be found in a variety of forest types, a study by Crowder & Taylor (1984) in Southern Ontario identified site characteristics associated with greater densities and biomass accumulation in this species. Shoot biomass was greater in sites with higher light intensities; however, it is thought that *M. canadense* can only occupy open sites if it is not limited by water stress. This species also occupied partially shaded sites but was not found in densely shaded areas. It occurs along a wide range of evaporation gradients but was found to have the highest concentration in humid sites. The presence of *Maianthemum* sp. was not related to soil depth, texture or pH. It was found to occur at medium levels of soil phosphorus availability. A study by McCall & Primack (1987) found this species to produce higher number of seeds and fruits when provided with nitrogen fertilizer. It has also been found to be resilient to metal stress and herbicide application (Gordon & Gorham 1963, Crowder & Taylor 1984) but cannot tolerate intense competition from sedges and grasses, which typically occupy disturbed sites that are dry and open (Crowder & Taylor 1984). Studies in the Sudbury region associated this species with undisturbed sites farther away from the smelters (15 to 35 km) (Rayfield *et al.* 2005). Based on vegetation surveys within this region this species can be found frequently throughout the Sudbury region and can be found at a closer distance to the smelters than *C.*

borealis (approximately 3 km), however it remains absent in areas that still remain affected by smelter emissions in the immediate vicinity of the smelters (Santala & Popp 2010, *unpublished data*).

Chapter 3

3 Methods

3.1 Study location

Geology

This study was conducted within the City of Greater Sudbury Ontario, Canada (46° 21’ N, 80° 59’W), approximately 390 km North of Toronto. This area was completely glaciated during the most recent glacial advance that occurred during the Pleistocene Epoch, which ended between 10,000 to 12,000 years ago. This area is located on the Southern Province and Grenville Province of the Canadian Shield and comprises sedimentary and volcanic rocks of the Huronian Supergroup together with intrusions from the Nipissing Gabbro. To the north of this region lies the Sudbury Basin, which is an elliptical structure (58 km long and 28 km wide) formed 1.85 billion years ago and is where nickel-copper sulphide ore bodies are found. Minerals of these ore bodies are, in order of abundance, pyrrhotite, pentlandite, chalcopyrite, magnetite and pyrite (Naldrett 1984; Rousell *et al.* 2002). The rim of the Sudbury basin ranges in elevation between 305 and 440 m above sea level. To the south of this rim, where this experiment was performed, ridges reach elevations of 300 m above sea level with differences in relief as great as 100 m. This area is characterized by glacial deposits of sand and gravel and thin discontinuous glacial till. These medium and course parent materials gave way to the development of podzolic soils that characterize study sites (Gillespie *et al.* 1983; Barnett & Bajc 1984).

Climate

The City of Greater Sudbury is located where the northern limit of the Great Lake St. Lawrence climate region meets the southern edge of the Northeastern Forest climate region (Environment Canada, 1998) with January temperatures averaging -13.5 °C and July temperatures averaging 19 °C. The monthly averages of the daily mean temperature are presented in Table 1. The total yearly precipitation is 899.3mm, 47% of which occurs during the months of May to September (Environment Canada, 2012). The average frost-free growing season ranges between 125 and 145 days (OMAFRA 2013). During the three years of this experiment, precipitation in 2010 was very low in comparison to historical average. Conversely in 2011, when the majority of my measurements were made, yearly precipitation had increased by 179 mm compared to the previous year; however, this was still below the historical average of 899.3 mm (Environment Canada, 2012). Yearly precipitation and growing degree days during the experiment are presented in Table 2. The predominant wind direction in this region commonly is from a south-westerly or north/ north-easterly direction (Watson *et al.* 2012).

Table 1: Monthly averages of the daily mean temperatures (°C) at the Sudbury Airport (46 37"N, 80 48"W, 347.5 m above sea level) in years 1971-2000 (Environment Canada, 2012).

Month	Daily Average (°C)
January	-13.6
February	-11.4
March	-5.3
April	3.1
May	11.3
June	16.2
July	19.0
August	17.7
September	12.3
October	5.8
November	-1.5
December	-9.5

Table 2: Yearly precipitation and growing degree days (above 5 °C) over three years of experiment based on data from the Sudbury Airport (Farmzone 2012, Environment Canada, 2012).

Year	Total Yearly Precipitation (mm)	Growing Degree Days
2010	650.8	1972
2011	829.1	1936
2012	758.2	2002

Vegetation

Forests in the Sudbury area are transitional between the Temperate Deciduous Forest to the South and Boreal Forest to the North (Scott 1995). Rowe (1959) and Braun (1964) describe this area as lying along the northern fringe of the Great Lake- St. Lawrence Forest Region. Records of the exact nature of vegetation that existed prior to industrial disturbance in this area are limited to land surveys that took place in 1856 and 1865 (Crown Land Department 1856; Canada Department of Crown Lands 1867). Drawbacks of these surveys were that they were limited to areas around canoe routes and exact distances travelled inland are not detailed. Also, identification of some tree species does not appear to be accurate. For example, the black and white oak identified along the Wanapitei River is more likely red oak or possibly bur oak. Despite these drawbacks, these surveys still provide a valuable picture of what the original forests of this area would have looked like. Vegetation surrounding Whitefish Lake, Round Lake and Mud Lake was described by Salter in 1856, in order of abundance, as being made up of timber birch, maple, white pine, red pine, hemlock, cedar, spruce, balsam, elm, ash and fine white oak. This area described by Salter would be approximately 3 km south of my most western study sites. In 1865, a surveyor by the name of Fitzgerald had a similar description and also noted that trees were enormous in size. He also described this area as having patches of hardwood, uplands, beaver meadows and cranberry marshes. In 1867 this surveyor described the vegetation along the Wanapitei River as having heavy timber that in many places had been destroyed by fire. He also identified white and blue oak (likely red oak) of average size along this river (Crown Land Department 1856; Canada Department of Crown Lands 1867).

Environmental damage

Since the late 1800's, the City of Greater Sudbury's landscape has been severely affected by the logging of timber, the mining of minerals, and the smelting of sulphate rich copper nickel ore (Winterhalder 1995). Based on findings from the Ontario/Canada Task Force (1982), over 100 million tonnes of sulphur dioxide and tens of thousands of tonnes of copper, nickel and iron have been released into the atmosphere as a result of over a century of mining and smelting activities within this region (Potvin & Negusanti 1995). The three main centres of ore roasting and smelting were located in Copper Cliff, Coniston and Falconbridge. A study by Murray and Haddow in 1945 found that the damaging effects of these emissions on tree foliage could be found as far as 35 km northeast, 20 km north and 20 km south of the smelters (Winterhalder 1995). Direct vegetation loss due both to industrial activities and to the associated environmental consequences of forest fire and increased frost action, have resulted in extensive soil erosion of the thin till of this region (Courtin 1994). With its black barren rocky hilltops devoid of vegetation, this denuded landscape was given the nickname the 'Sudbury Moonscape' by the media (Winterhalder 1995; Lautenbach *et al*. 1995).

Restoration effects and recovery

Enhanced smelter technology and increased government air pollution controls have reduced the amount of industrial emissions and have allowed Sudbury's disturbed landscape to slowly recover (Potvin & Negusanti 1995, Watson *et al.* 2012). Emission reductions resulting from smelting facility upgrades are provided in Table 3 (Watson *et al.* 2012).

Table 3: Total Annual Emissions (tonnes $year^{-1}$) from Sudbury Area Smelters (Environment Canada 2010 from Watson *et al.* 2012).

Smelter	Year	Cu	Ni	Pb	Co	As	SO_2
Sudbury INO Falconbridge Smelter	2009	7.2	13	5.1	1.1	0.83	32,714
	1995	9.1	12	14	0.71	na	na
Vale Copper Cliff Smelter	2009	42	14	8.1	0.28	2.4	41,993
	1995	107	418	68	5.6	7.3	236,033

Table 4: Summary of mean concentrations ($mgkg^{-1}$) of select metals in surface soil (0-5 cm depth) in residential areas in the City of Greater Sudbury. Ministry of the Environment Standards were taken from the Generic Site Condition Standards for shallow soils in potable ground water conditions for residential/parkland land use (SARA Group 2009, OMOE 2011). Community Groupings for this study are presented in Figure 1.

Metals	As	Co	Cu	Pb	Ni	Se
Ministry of the Environment Standards	18	22	(180) 140	120	(130) 100	2.4
Copper Cliff	18	33	1440	91	1017	7.8
Coniston	10	16	246	52	336	1.2
Falconbridge	74	56	874	88	956	2.7
Sudbury Core	10	18	392	67	400	1.0
Inner Sudbury	5.5	9	106	23	124	0.7

( ) Standard in parentheses applies to medium and fine textured soils

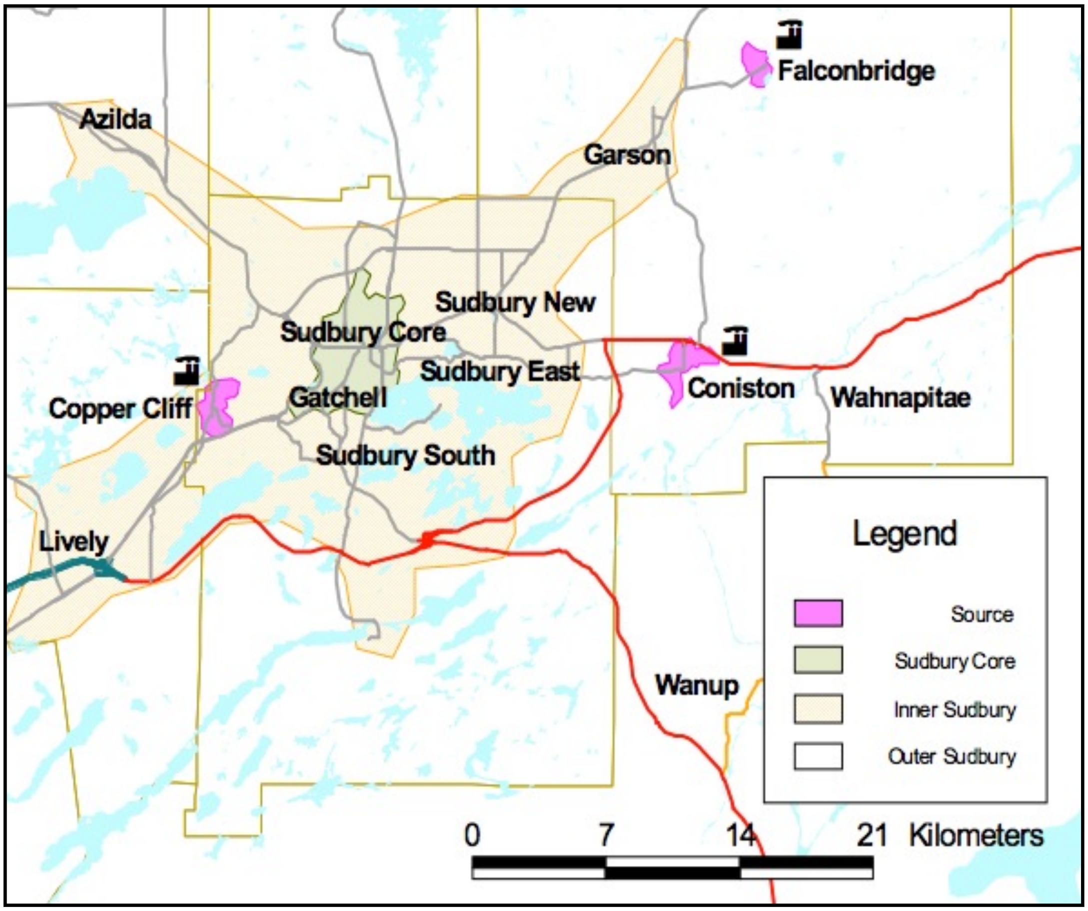

Figure 1. Community groupings for Sudbury soil study sampling (OME 2004).

Landscape recovery has been augmented through municipally and industrially driven reclamation initiatives. These activities included the application of limestone and fertilizer to increase soil pH, reduce metal availability and increase soil nutrient availability. Seeding with agronomic grasses was used to stabilize soils and create a favourable microclimate for natural colonization of birch, poplar and willow and planted coniferous trees. Since the beginning of the reclamation program in 1978 until 2012, over 9.3 million trees and over 125 thousand shrubs have been planted and close to 3, 500 hectares of land have received limestone within the region (City of Greater Sudbury 2012a, City of Greater Sudbury 2012b). Primarily, reclamation work occurred in areas adjacent to major roads and urban neighbourhoods. As a result of these actions decades ago, seedlings have grown into what appears to be a young forest (Lautenbach *et al.* 1995; City of Greater Sudbury 2012b).

Amiro & Courtin (1981) described nine major plant community types within the Sudbury region in the late 1970's. Three communities were affected by pollution and consisted of barren, birch transitional and maple transitional communities. The other six include birch-maple community, red oak community, poplar lowland community, jack pine community, largetooth aspen community and sugar maple community. Sinclair (1996) who described vegetation communities again in 1993 found six plant community types within the Sudbury area. These communities, named after the dominant tree species making up their canopy, include the birch/pine (red and jack pine), trembling aspen, red oak, big-toothed aspen, and white birch communities. A tree-less canopy community comprised of *Agrostis scabra* and *Deschampsia caespitosa* was also recognized. Sinclair noted that Amiro and Courtin's red maple community was absent. Its disappearance was related to the retrogressive dieback of this species (Sinclair 1996).

Even though reclamation has changed large areas once completely devoid of vegetation to what now looks like young forests, the recovery of this area is still not complete. The biodiversity action plan for the City of Sudbury estimates that 30 thousand hectares have yet to be planted with trees or to receive limestone. Moreover, succession of understory species within this region appears to be delayed so initiatives to enhance biodiversity have been employed such as the planting of understory shrubs and herbs (Braun & O'Hara 2006 *unpublished data*; City of Greater Sudbury 2012b). An ecological risk assessment has also revealed that terrestrial plant communities within this area continue to be affected by elevated soil levels of arsenic, cadmium, cobalt, copper, nickel, lead, selenium (Table 4), low soil pH, soil erosion, and lack of soil organic matter (SARA 2009).

3.2 Planting and field methods

Transplanting technique for the forest floor mats

Between April and September of 2010, forest floor plants were transplanted by the City of Greater Sudbury Land Reclamation Department. The date plots were transplanted and weather conditions on those dates are included in Appendix A. Low shrubs and herbaceous species typical of Ontario pine forests were deemed appropriate for introduction based on recommendations by Braun (2007). Forest floor plants were salvaged from an area slated for the expansion of Highway 69, located 50 km south of Sudbury, hereafter referred to as the donor site (from N 46°10.334’, W 80°43.011’ to N46°09.243’, W 80°42.517’). Donor sites were located in areas that would not have experienced vegetative damage resulting from smelter emissions (Murray & Haddow 1945, as cited by Winterhalder 1995). Vegetation was removed in mats 64 cm long, 56 cm wide and 10 cm thick (25” long, 22” wide and 4” deep) using shovels. The mat was transferred into a plastic bread tray of the similar dimensions (Figure 2). The rational for using a 10 cm thick matt was that it allowed majority of root system to be harvested. This thickness also prevented aboveground plant material from being crushed when plastic bread trays were stacked one on top of each other during transport. The material was transported out of the field using ATVs and transported back to Sudbury using a moving van (900 cu. ft and 4, 000 lb load capacity). This material was then transported to an outdoor storage facility in Sudbury where trays were unloaded, stacked, watered and covered with a white fabric sheet to prevent desiccation. Plants only received water at this outdoor facility and did not receive additional watering following transplantation as weather conditions during the summer of 2010 did not warrant it.

Material was transplanted into 16 m^2 plots (4m by 4 m), hereafter referred to as receptor sites, one to three days following collection. The decision to use large 16m^2 plots was decided by the Vegetation Enhancement Technical Advisory Committee with the rationale that if edges of plots dried out due to desiccation of this exposed edge, plants within the plot would be protected. In total, 30 transplant plots were observed in my investigation. Transplant plots were distributed across a gradient of metal contamination (Figure 4) in areas with canopy cover ranging from 50% to 100%. Receptor plots also varied in soil and habitat types. Prior to transplanting, the plots were prepared by removing any leaves or sticks with a rake. The substrate was also loosened with a garden fork to a depth of approximately 4 inches. Forest floor mats were then placed within the 16 m^2 plot such that edges of the individual mats were flush with one another to minimize desiccation within the plot. Great care was taken to ensure that plots were exactly 16 m^2. Galvanized steel posts and nails were placed in corners of plots to guarantee that the original size of plot could be determined in the future (Figure 3). After the forest floor mats were placed into the plot, leaves were raked up along the plot edges of the mats to prevent edges from drying or eroding. In 2011, six quadrats (16m^2 in area) were marked off in undisturbed areas of the donor site to serve as a comparison of environmental and growth characteristics between experimental plots and undisturbed vegetation in the donor site.

Figure 2. Understory material being salvaged from donor site.

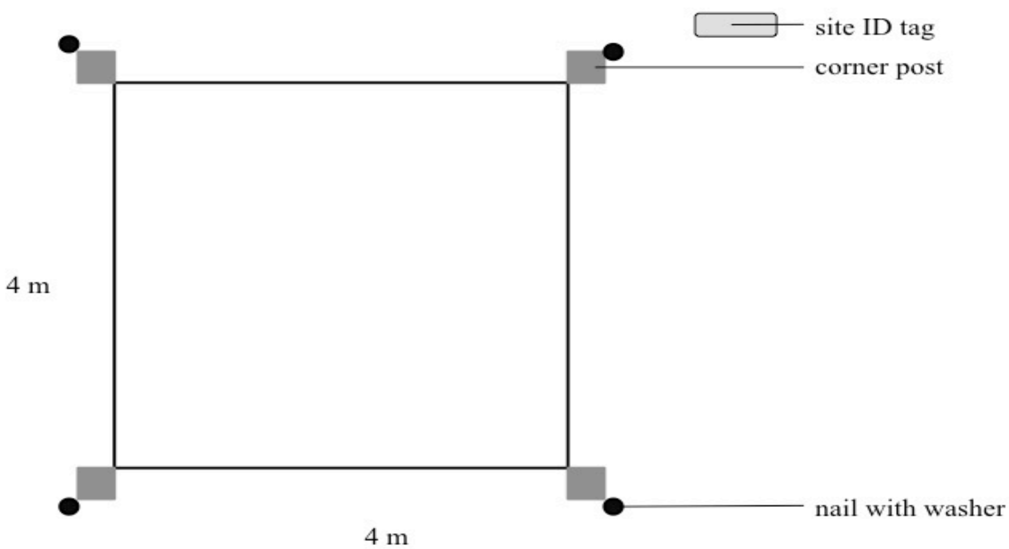

Figure 3. Plot design.

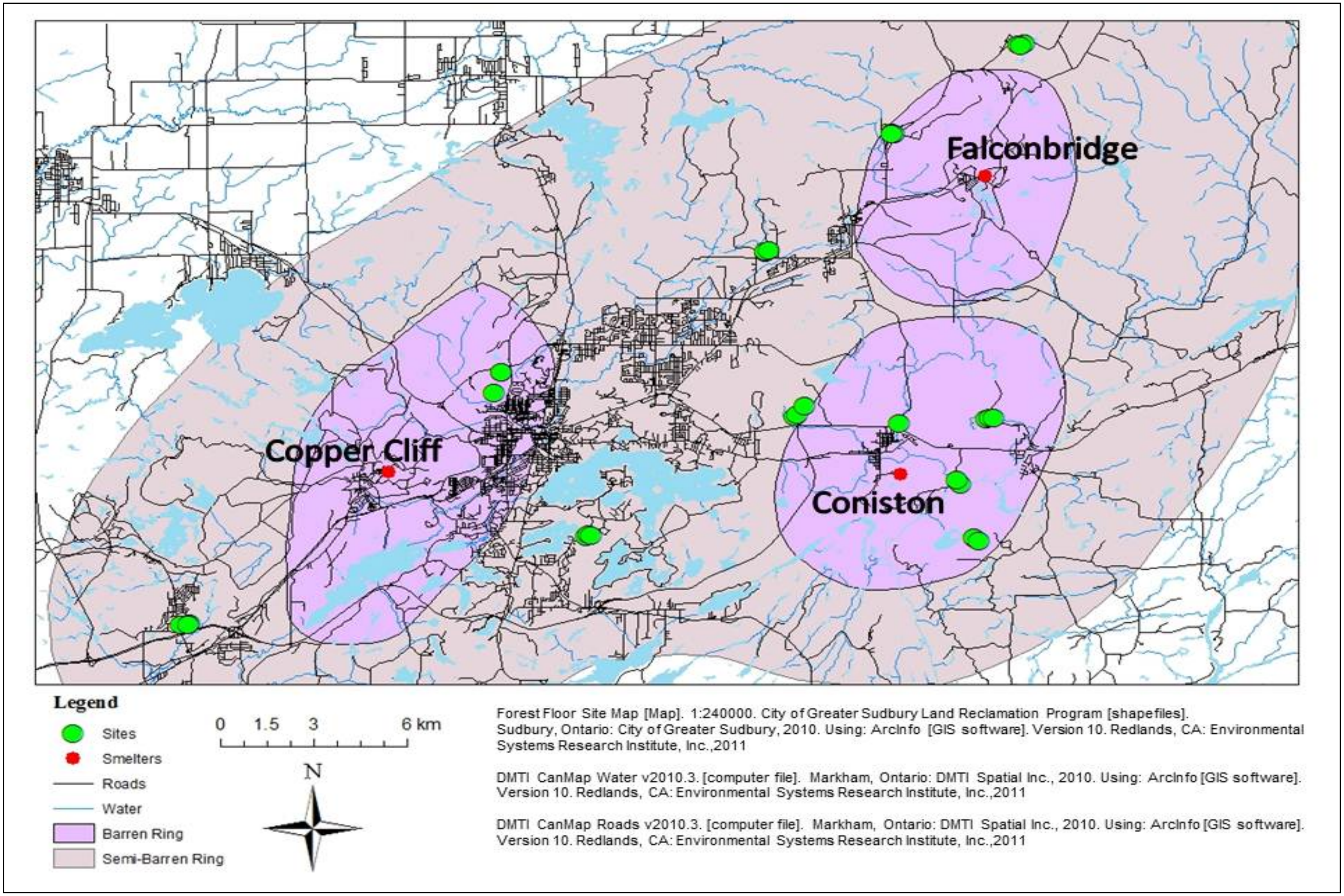

Figure 4. Location of study sites in relation to Struik's (1973) barren (devoid of trees and sparse of vegetation) and semi-barren (transition between the barrens and the natural plant community of the region) (Amiro & Courtin 1981) vegetation zones within the City of Greater Sudbury. Operating smelters are located in Copper Cliff and Falconbridge, where the Coniston smelter operated until 1972 (Winterhalder 1995).

3.3 Evaluation of transplant success

Evaluation of Transplant Success: Root Ingrowth Cores

To compare root growth in recipient site soils and donor site soil between the plots, root ingrowth cores (20 cm in depth and 5 cm in diameter) were installed between 19 May and 21 June 2011. Ingrowth cores were placed between two individuals of blue bead lily (*Clintonia borealis* (Aiton) Raf.) and between two individuals of wintergreen (*Gaultheria procumbens* L.) (Figure 5). I chose these species primarily because they were present in every one of my research plots. These species were also chosen because they can be found occasionally throughout the Sudbury region and indicating that they are able to tolerate some metal stress. This fact made them ideal candidates for this assessment, as they would possibly respond to polluted conditions by reducing growth rather than dying. More vulnerable species might just die on the most polluted sites. As well, physiologically and ecologically these two species differ. *C. borealis* is a shade-tolerant herb with deep roots (Carter & Vogelmann 1968; Ashmun & Pitelka 1985; Pitelka *et al.* 1985; Utech 2008). In contrast, *G. procumbens* is a sun-tolerant evergreen shrub with roots that are restricted to the litter layer (Donohue *et al.* 2000; Roberts & Lixiang 2002; Moola & Vasseur 2009; Trock 2009). This species is also tolerant of ecological changes associated with disturbances such as canopy removal and has an affinity for acidic soils (Moola & Vasseur 2004; Moola & Vasseur 2009).

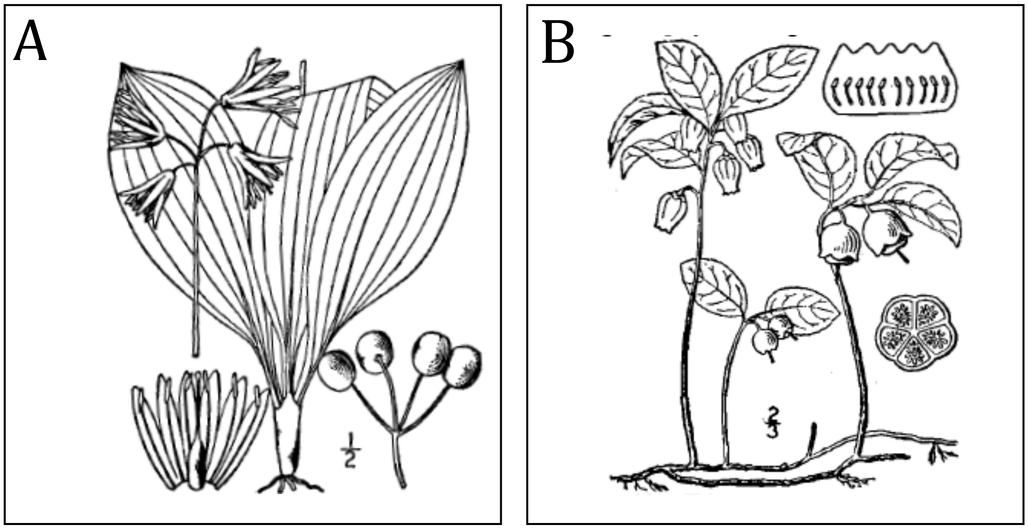

Figure 5. A. Blue bead lily *Clintonia borealis* (Aiton) Raf B. Wintergreen *Gaultheria procumbens* L. (from Britton &Brown 1943).

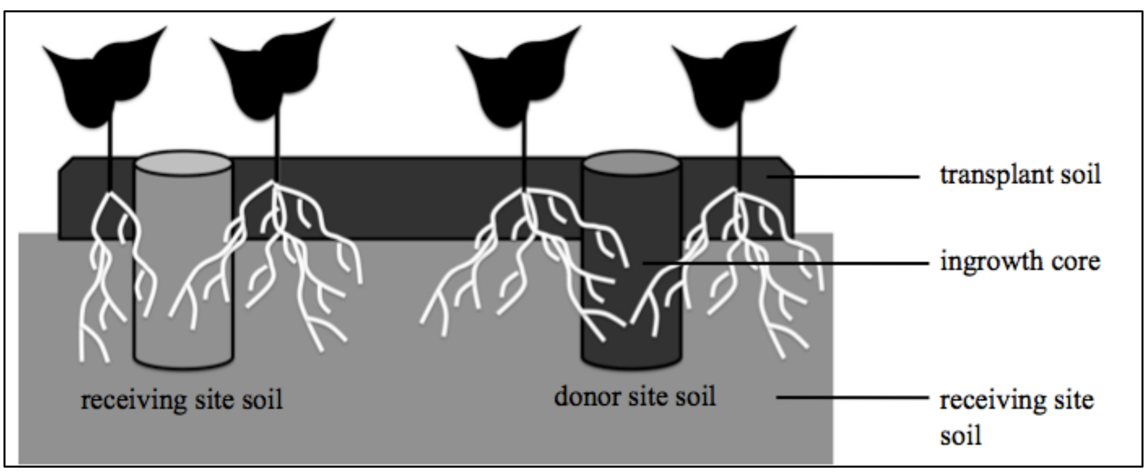

Figure 6. Placement of ingrowth cores for each species.

On each plot two ingrowth cores were placed between two individuals of the investigated species (Figure 6). One ingrowth core was filled with the donor site soil, while the other was filled with receptor site soil such that there were two cores per species per plot. Receptor soil cores were filled with soil collected from a distance of 1 metre around each plot so that soil collected corresponded to the location of each individual plot. Donor cores were filled with soil collected from two undisturbed areas located near the donor site, 3 km from each other. Soil was collected from the top 5 cm soil layer for both donor and receptor site soils because the SARA-Group (2001) study found that this top 5 cm layer in Sudbury had elevated concentrations of metals and lower pH. The collected soil was passed through a 1 cm sieve to remove any roots.

Ingrowth cores were installed by removing cylindrical cores of soil 20 cm in depth and 5 cm in diameter from within the plot using a stainless steel auger (CM. Nelson Welding). A plastic tube slightly smaller than the diameter of the hole was then placed into a tubular nylon sock (5 mm mesh size) with the same dimensions as the removed core, and both were placed into the hole. The nylon sock was then filled with sieved root-free soil. To achieve the same bulk density within each core, the soil was added in 2 cm layers at a time and then lightly packed with a wooden dowel. This process was repeated until the ingrowth core was filled. Two ropes attached to the bottom of the nylon mesh bag were left above the soil surface to aid in the removal of cores at the end of the season. Ingrowth cores were removed after 11 weeks and were stored at 5 ^{0}C until the roots were measured one to five days later. Any roots outside the mesh core were trimmed flush to the core. Soil was washed from the roots, which were then dried in a drying oven at 72^{0}C for at least 48 hours then measured for dry mass. Root mass density (mg_{root}/cm^{3}_{soil}) was then calculated as dry mass of the roots divided by the volume of the core.

Evaluation of transplant success: sexual reproduction and vegetative spread

To assess establishment and performance of plants in the transplants, the number of fruits and flowers produced by three of the species, Canadian mayflower (*Maianthemum canadense* Desf.), bluebeaded lily (*Clintonia borealis* (Aiton) Raf.) and wintergreen (*Gaultheria procumbens* L.) were measured for each plot as a measure of sexual reproduction. These species were primarily chosen because they were the only three species common to all plots observed in this study, and because they have different reproductive strategies. *M. canadense* Desf. is self-incompatible and requires insects to pollinate it (Worthen & Stiles 1986; Hisatomo *et al.* 2008). *C. borealis* (Aiton) Raf.is capable of self-pollination and cross-pollination; however, seedlings of this species are rarely seen (Angevine & Handel 1986; Dorken & Husband 1999). *G. procumbens* L. is considered to be self-fertilizing, but is also a facultative out-crosser and is pollinated by insects (Sobey & Barkhouse 1977; Flinn & Pringle 1983; Donohue *et al.* 2000). The number of fruits and flowers produced by each individual of these three species within the plots was recorded in early July 2011 and in early July again in 2012. As a measure of vegetative spread, the distance of lateral spread of each species producing runners that emigrated beyond the edges of the plots was measured on 18 July 2011. Cover and abundance of all the species within each plot were determined by using a modified Braun-Blanquet cover scale (Table 5) on 25 June to 11 July 2011.

Table 5: Modified Braun-Blanquet Cover Scale (Elzinga *et al.* 1998)

Cover Scale	
5	75% - 100%
4	50% - 75%
3	25% - 50%
2	5% - 25%
+	less than 5%
r	a single plant

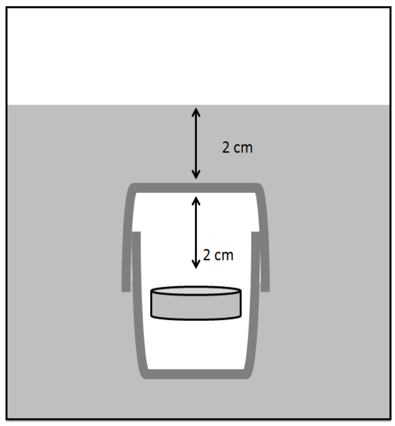

Figure 7. Placement of temperature data logger below soil surface (light grey).

3.4 Environmental site characteristics

Microclimate

The percent cover of overstory above the plots was measured from 30 June 2011 to 7 July 2011 using a spherical densiometer (Lemmon 1956). The maximum, minimum, and average daily temperature, average change in diurnal temperature and soil growing degree days were measured for each plot using temperature data loggers (Maxim iButton® DS1921G-F5#, San

Jose, California, USA) placed 4 cm below the soil surface in a PVC waterproof container (3.6 cm in height and 3 cm wide) in the centre of each plot (Figure 7). The data loggers measured the temperature every 255 minutes from 18 July 2011 to 27 June 2012. Soil growing degree days (equation 1) (OMAFRA 2011) were calculated using a base temperature of 5 °C because previous studies used base temperatures of 5 °C for *Anemone nemorosa* L., 4.4 °C for *Trientalis borealis* Raf. and 5°C for temperate forest herbs in Northern Europe were used to calculate growing degree days (Anderson & Loucks 1973; De Frenne *et al.* 2011) .

$$\text{Soil Growing Degree Days} = \sum \frac{\text{Max. Temperature} + \text{Min. Temperature}}{2} - \text{Base Temperature} \quad \text{(Equation 1)}$$

The accuracy of the temperature readings of the iButton® datalogger in comparison to temperature readings taken using 5 constantan copper-nickel thermocouple logged using an 8 channel datalogger (21 X Micrologger, Campbell Scientific, USA) was tested in a separate experiment. This experiment occurred over a 24 hour period (midnight until midnight) on 15 June 2013. Readings of the iButton® datalogger were more similar to readings of the thermocouple placed at a depth of 0.5 cm below the soil (Figure 8). The iButton® readings were also warmer in the evening and cooler during the day in comparison to those of the thermocouple placed 0.5 cm below the soil surface. This accounted for an average difference in temperature readings of 0.6 º C. As a result the ibutton® datalogger measured a soil growing degree day value that was 0.6 units higher than that calculated by data collected by the thermocouple 0.5 cm below the soil surface. Therefore, soil growing degree day readings taken with the ibutton® are 0.6 º C higher than that taken with a thermocouple at 0.5 cm below the soil surface. However, based on these findings I do not suspect differences in rank order of the plots for soil growing degree days.

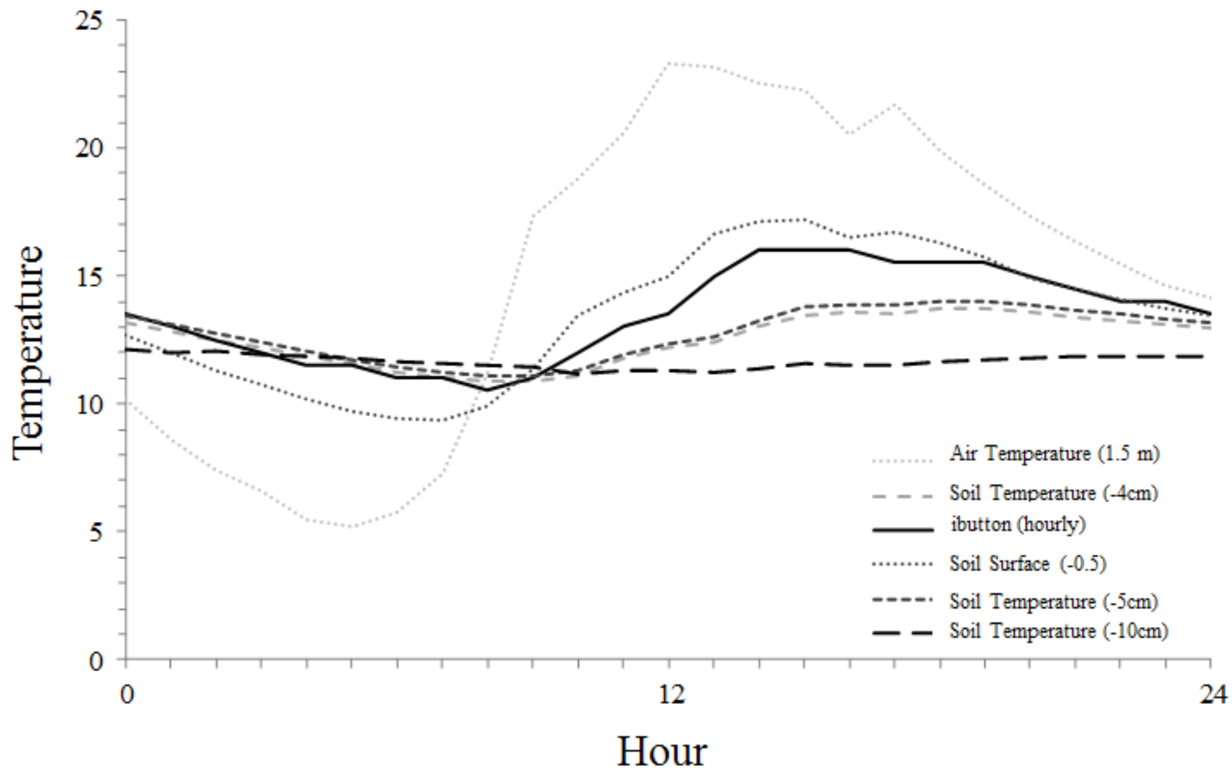

Figure 8. Soil temperature ºC within plots on 15 June 2013 recorded from 12 midnight (0 hour) until 12 midnight of the following evening (24 hour). An 8 channel datalogger recorded temperature of the air (1.5 cm above soil surface) and at depths -0.5, -4cm, -5cm, and -10 cm below the soil surface (hatched lines). Ibutton® recorded every hour during the same time period (solid lines).

Soil Chemical Analyses

In May of 2012, soil samples were taken at a distance spanning 1 metre from the receptor and donor plot edges. In total 18 soil cores per plot (5cm in length and diameter) were collected using a soil corer (Nelson GM Ltd., Sudbury Ontario) and all were pooled into a single sample. The decision to have a single pooled sample per plot was based on financial consideration of the soil sampling budget. For donor soil, samples of soils were taken from buckets used to store donor soil. In total, three samples of donor soil were analyzed, each of which comprised subsamples randomly selected from all storage buckets. To determine plant-available elements content in the underlying substrate, air-dried samples were extracted using a 1 M ammonium acetate solution (Baker *et al.* 1994) and analyzed with inductively coupled plasma mass spectroscopy (ICP-MS, Testmark Laboratories, Sudbury Ontario). Strong acid extractable elements were analyzed using a 1:1 nitric acid:hydrochloric acid extraction and then analyzed with inductively coupled plasma mass spectroscopy (ICP-MS, Testmark Laboratories, Sudbury Ontario). Plant available phosphorus was determined using a sodium bicarbonate extraction (Olsen 1954), and potassium, calcium and magnesium were determined using an ammonium acetate extraction (Agrifood laboratories, Guelph, Ontario). Total organic carbon was analyzed by using an oxidative/combustive process that measured the amount of carbon dioxide liberated by the sample using non-dispersive infrared techniques (Walkely & Black 1934). Nitrogen in the soil was measured as nitrate-nitrogen and ammonium (Agrifood laboratories, Guelph, Ontario). Soil pH was measured using saturated paste method while Bph was tested using SMP buffer pH method (Agrifood laboratories, Guelph, Ontario).

3.5 Statistical analyses

Statistical analyses were performed using SPSS version 19. The root mass density was first analyzed using a split-split plot analysis with smelter affected zone (barren or semi-barren) as the main plots factor, and plant species (*C. borealis* and *G. procumbens*) in the first split-plots and soil types (donor or receptor site soil) in the second split-plots. Smelter affected zone was considered a random effect whereas soil and species were considered as fixed effects. Root mass density was square-root transformed to meet the normality assumptions of the analysis. Data from plants grown at the donor site were not included in the statistical analysis.

Multiple linear regression analyses were conducted to identify which environmental factors best predicted (i) root mass density for *C. borealis* or *G. procumbens*; (ii) the frequency of flowering of *G. procumbens*; (iii) the number of plants with fruits for *M. canadense* or *C. borealis*; (iv) the number of target species spreading from the plots; (v) species richness per plot; (vi) Shannon-Wiener diversity index per plot; and (vii) species evenness per plot. Non-target species were not considered in calculation of species diversity. These species include *Hieracium aurantiacum* L., *Hieracium caespitosum* Domort., *Lotus corniculatus* L., *Fallopia cilinodis* (Michx.) Holub, *Rumex acetosella* L., *Taraxacum officinale* F.H. Wigg., any *Solidago* sp. L.. These species originated either as an unintended introduction from the donor site or originated from the receptor site and colonized the plots naturally.

Before conducting multiple regressions, the number of independent variables was reduced using a three-step strategy to reduce the potential for collinearity in the regression analyses. First, a full correlation matrix was used to determine the potential for correlated variables. Arsenic, selenium, lead, cobalt, nickel, organic matter content, phosphorus, potassium, magnesium, calcium, sulphate, nitrate nitrogen and ammonium nitrogen were log transformed because they were highly skewed and consequently had excessive leverage which would violate assumptions of regression analysis (Quinn & Keough 2002). One severe outlier from percent canopy cover data set was detected using Grubbs' test and removed (p<0.05). For the second step, two groups of related independent variables were each subjected to a principal component analysis (PCA) with varimax rotation to choose key variables. These groups consisted of plant-available chemicals of concern (As, Cu, Co, Ni, Pb and Se) and plant-available nutrients (NO_3^-, NH_4^+, P, K, SO_4^{-2}). The suitability of the data for the PCA was assessed using the Kaiser-Meyer-Olkin measure of sampling adequacy (> 0.6) and Bartlett's test of sphericity ($p < 0.05$; Norman & Streiner 2008). Eigenvalues and scree plots were used to determine principal components. Independent variables with the highest loading values from these rotated principal components were then retained for evaluation for collinearity. For the third step, collinearity of all remaining independent variables was then evaluated using Pearson product-moment correlation coefficients and collinear variables ($P < 0.05$) were then selectively removed.

Stepwise multiple regressions were then conducted for each dependent variable using the remaining independent variables, and best models were selected using Akaike information criterion (AIC). To meet assumptions of normality and homogeneity of variance for regression analysis, the root mass density for each species was square-root transformed and the number of fruiting *M. canadense* individuals was log transformed.

Chapter 4

4 Results

4.1 Split-split plot analysis of root mass density

Root mass density was not affected by the smelter-disturbance zone, the species, or the type of soil in which they were grown, as determined by the split-split plot ANOVA (Table 6, Figure 9). There were also no significant interactions among any of the three independent variables. However, within the barren zone, the mean root mass density of *C. borealis* and *G. procumbens* grown in the receptor site soil was 25% and 19% smaller, respectively, when compared to root growth of the same species grown in donor site soil, but this interaction was not statistically significant (p = 0.129). No such pattern was seen within the semi-barren zone.

Table 6: General linear model of split-split plot analysis on effects of smelter distance, soil type and species root mass density (mg_{root}/cm^3_{soil}). Sites were considered as a random factor, smelter affected zone (barren or semi-barren) as the main plots factor, plant species (*C. borealis* and *G. procumbens*) in the first split-plots and soil types (donor or receptor site soil) in the second split-plots.

Source	df	MS	F	*p*-value
Smelter zone	1	0.198	1.871	0.182
Error A	28	0.106		
Species	1	0.198	1.836	0.187
Species × Smelter zone	1	0.004	0.078	0.782
Error B	27	0.108		
Soil	1	0.031	1.155	0.287
Smelter zone × soil	1	0.065	2.379	0.129
Species × soil	1	4.51×10^{-5}	0.002	0.968
Smelter zone × species × soil	1	0.008	0.299	0.587
Error C	56	0.027		
Total Error	118			

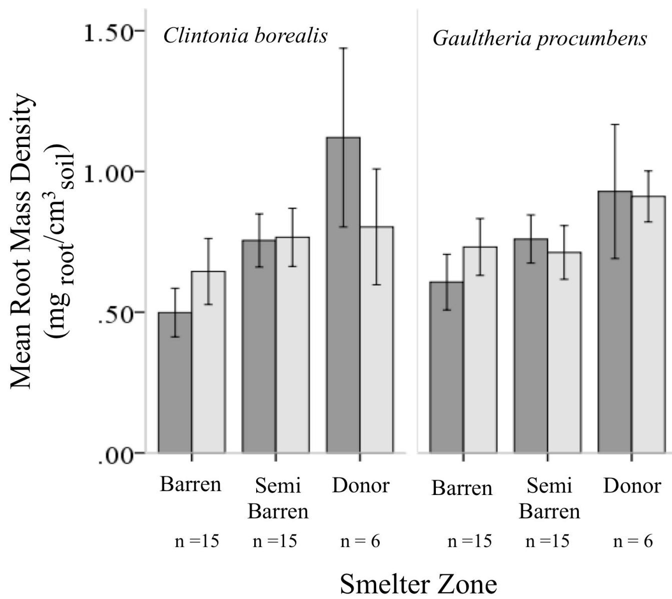

Figure 9. Root mass density (mg_{root}/cm^3_{soil}) of *C. borealis* and *G. procumbens* grown in receptor site soil (dark grey) and donor site soil (light grey) on sites located within the barren zone, semi-barren zone and donor site (mean ± 1 S.E.). Donor zone data were not included in the statistical analysis.

4.2 Multiple regression analyses of site characteristics on transplant establishment

Pearson's product moment correlations were used to determine the potential for environmental characteristics to be correlated with each other (Appendix B). Descriptive statistics are presented in Appendix E, F and G. Soil growing degree days (soilGDD), percent canopy cover, distance from the smelter, organic matter content, BpH, cation exchange capacity (C.E.C) were retained as independent variables for regression analysis. BpH was used as a predictor variable instead of soil pH because pH could not be transformed to attain assumptions of normality for regression analysis, and both these variables were strongly positively correlated ($r=0.9$, $p<0.05$). Other variables related to plant available chemicals of concern, and plant available nutrients were subjected to a principal components analysis (PCA) to reduce the number of variables for regression analysis.

For chemicals of concern, three principal components had eigenvalues exceeding 1, explaining 50.4%, 26.2%, and 16.8 % of the variance, respectively. The first component revealed strong positive loading for arsenic and lead ($r > 0.9$) and the second component had strong positive loadings for nickel and cobalt ($r > 0.9$; Figure 10). Only arsenic and nickel were retained for multiple regression analyses. The principal components analysis of the seven variables relating to plant-available nutrients (nitrate nitrogen, ammonium nitrogen, calcium, magnesium, sulphates, potassium and phosphorus) gave two principal components with eigenvalues exceeding 1, explaining 45.8%, 18.8% of the variance respectively. Rotated components matrix for the first principal component revealed strong positive loading for calcium ($r > 0.8$) and negative loadings for phosphorus ($r < -0.6$; Figure 11). The second principal component showed strong negative loadings for sulphates ($r < -0.7$) and strong positive loadings for both ammonium and nitrate nitrogen ($r > 0.6$). Phosphorus, sulphates, as well as ammonium and nitrate nitrogen, summed as total inorganic nitrogen, were retained for multiple regression analysis.

A strong correlation was found between C.E.C and BpH ($r = 0.95$), which violates assumptions of regression analysis (Appendix B). Therefore, C.E.C was dropped from the analysis. The independent variables retained for regression analyses consequently consisted of soil growing degree days, percent canopy cover, plant available nickel, arsenic, phosphorus and sulphates, total nitrogen, distance of the plot from the closest smelter, BpH, and organic matter content. Stepwise multiple regressions were conducted for each dependent variable using these retained independent variables, and best models were selected using Akaike information criterion (AIC).

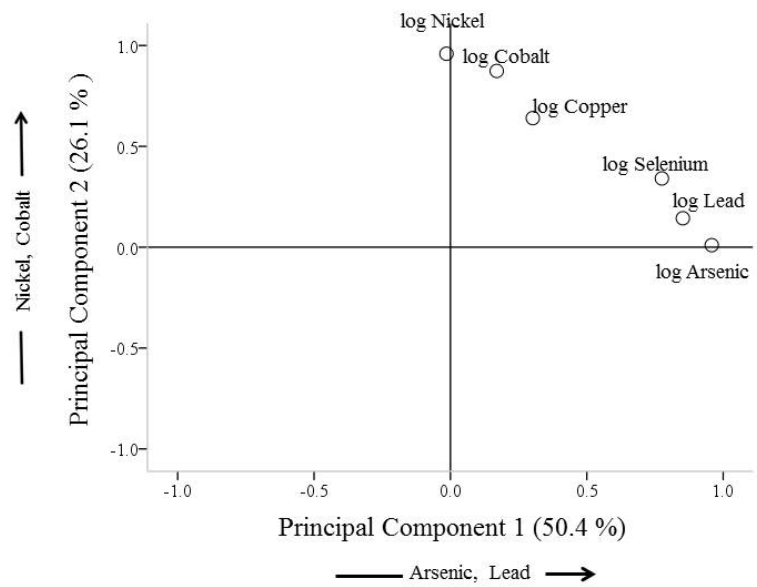

Figure 10. Rotated principal component plot of plant available chemicals of concern.

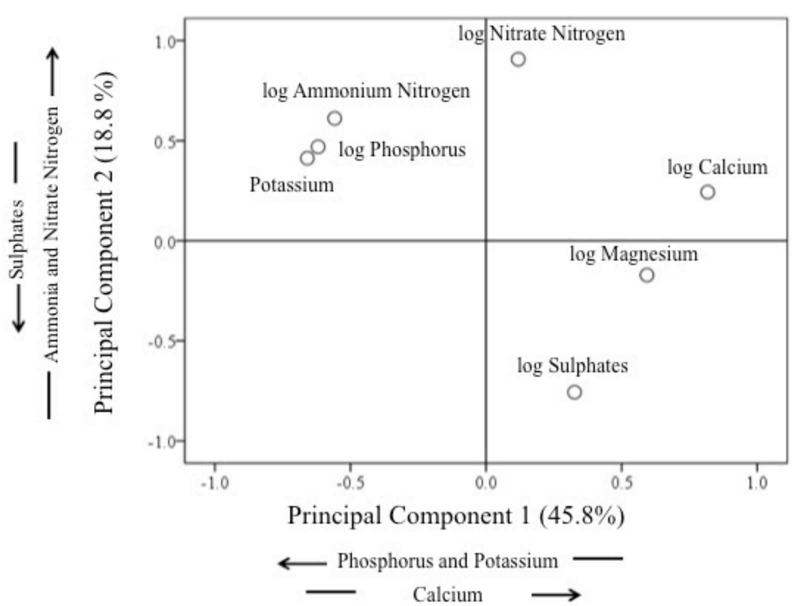

Figure 11. Rotated principal component plot for plant available nutrients

Stepwise multiple regression found that the root mass density of *G. procumbens* and *C. borealis* respond to different environmental variables. The rooting density of *G. procumbens* was best predicted by plant available nickel (unstandardized Beta = - 0.43, $p = 0.001$) in soils surrounding the plot and soil growing degree days (unstandardized Beta = 0.001, $p < 0.01$) (Table 7). Root growth was negatively affected by plant available nickel and positively affected by soil growing degree days. In contrast, the root density of *C. borealis* was not affected by smelter-related variables and was best predicted only by soil growing degree days (unstandardized Beta =0.001, $p < 0.05$) (Table 7); root mass density of this species also increased in plots with warmer soils. The model with both soil growing degree days and log total nitrogen was not selected because nitrogen (unstandardized Beta = 0.16, $p = 0.25$) did not make a significant contribution to that model. Best-fit regressions for these models selected using AIC for root growth in *G. procumbens* and *C. borealis* are presented in Table 8.

Flowering frequency of *G. procumbens* was best explained by another variable, BpH (unstandardized Beta = -2.42, $p < 0.05$) in addition to percent canopy cover (unstandardized B = 0.11, $p < 0.05$) (Table 7). Plots with high BpH had a low frequency of flowering individuals and plots with greater canopy cover had a higher flowering frequency. The number of fruiting individuals of *C. borealis* was negatively related with sulphates (unstandardized Beta = -4.51, $p < 0.05$) within the receptor site soil but positively related with plant available nickel (unstandardized Beta = 2.31, $p < 0.05$) (Table 7). For the number of fruiting individuals of *M. canadense*, the distance of the plot from the smelter (unstandardized Beta = - 1.06, $p < 0.05$) and plant available phosphorus (unstandardized Beta = -0.45, $p < 0.05$) were highly significant predictors in the multiple regression (Table 7). Number of flowering individuals was postively related to distance away from the smelter and negatively correlated with plant available phophorus. Best-fit regressions for these models for flowering frequency of *G. procumbens* and number of flowering individuals of *C. borealis* and *M. canadense* are presented in Table 8.

Table 7: Model explaining variance of dependent variable. The best model based on AIC in bold.

Dependent variable	Model[1]	r^2	AIC	*F*	*P*
Roots *Gaultheria*	log Ni	0.35	-88.6	14.3	0.001
	log Ni + sGDD	**0.48**	**-92.8**	**11.7**	**<0.001**
Roots *Clintonia*	**sGDD**	**0.17**	**-83.5**	**5.1**	**0.034**
	sGDD + logTIN	0.21	-83.0	3.3	0.056
Flowers *Gaultheria*	BpH	0.21	47.0	7.0	0.014
	BpH + % cover	**0.33**	**44.5**	**6.1**	**0.007**
Fruits *Clintonia*	logSO_4^{2-}	0.19	31.9	6.1	0.021
	logSO_4^{2-} + log Ni	**0.32**	**28.9**	**5.8**	**0.008**
Fruits *Maianthemum*	log distance	0.22	-58.5	7.4	0.011
	log distance + log P	**0.36**	**-61.9**	**6.9**	**0.004**

[1]Independent variable in model: plant available nickel (Ni), soil growing degree days (sGDD), total inorganic nitrogen (TIN), buffering pH (BpH), percent canopy cover (% cover), plant available sulphates (SO_4^{2-}), plant available phosphorus (P), and distance from the smelter (distance).

Table 8: Best-fit regression models explaining variance of dependent variable.

Dependent variable		n	Best - fit regressions[1]
Root	*Gaultheria*	28	$y = -0.43\ \mathbf{x_{Ni}} + 0.001\ \mathbf{x_{sGDD}} + 0.55$
	Clintonia	30	$y = 0.001\ \mathbf{x_{sGDD}} + 0.24$
Fruits	*Gaultheria*	30	$y = -2.42\ \mathbf{x_{Bph}} + 0.11\ \mathbf{x_{\%canopy}} + 8.27$
	Clintonia	30	$y = -4.51\ \mathbf{x_{logSO4^-}} + 2.31\ \mathbf{x_{logNi}} + 6.19$
	Maianthemum	30	$y = 1.06\ \mathbf{x_{logdistance}} - 0.45\ \mathbf{x_{logP}} + 0.47$

[1]Independent variable in model: plant available nickel (Ni), soil growing degree days (sGDD), buffering pH (BpH), percent canopy cover (% cover), plant available sulphates (SO_4^{2-}), plant available phosphorus (P), and distance from the smelter (distance).

Environmental site characteristics did not explain variation in spread of species from the plot, the number of non-target species (Appendix C), Shannon-Wiener Diversity Index, evenness or species richness within the plots (Appendix D). The number of individuals that spread from receptor plots ranged between 0 and 26 individuals, and on average 3.5 individuals spread from these plots. Twenty-one receptor plots had non-target species within them, and on average 1.2 non-target species were found within the plots. Shannon-Wiener Diversity Index was 2.65 for receptor plots and 2.4 in donor site plots. Although variation in species evenness within the plot was weakly predicted by distance of the plot from the smelter ($F_{(1,28)}$ = 3.70, r^2=0.12, p=0.065), the range in species evenness within transplant plots (between 0.67 to 0.96) does not appear different from that of the donor sites (between 0.72 to 0.93). Species richness was higher in receptor plots than donor sites such that there were 23.5 species on average within receptor plot sites and 17.2 species on average within donor plot sites.

Chapter 5

5 Discussion

When using transplanted vegetation in ecosystem restoration, the placement of plants into sites with suitable microclimate is essential for successful reintroduction (Mottl *et al.* 2006; Fahselt 2007; Drayton & Primack 2012). In this study I found that environmental variables of the transplant site had an influence on transplant success and that predictors were either related to smelter emissions or other natural environmental variables, and that these results were species specific. I also found that root growth of species in receptor site soil was the lowest in sites with the highest metal contents; however, differences in root growth of both species grown in the barren zone located closest to the smelter were not significantly different than those grown in the semi-barren zone farther away. The application of these findings to ecological restoration will be discussed at the end of this chapter.

5.1 Root growth

One question that was asked in this investigation was whether transplant roots would be capable of growing into the receptor site soil because root growth in organic amendments has been found to stop once roots come in contact with metal-contaminated substrates below (Johnson *et al.* 1977). Elevated concentrations of Cu and Ni are known to cause significant reductions in root growth in *Leucanthemum vulgare* Lam., *Fragaria vesca* and *Arctostaphylos uva-ursi* (Salemaa & Monni 2003; Roiloa & Retuerto 2006; Ryser & Emerson 2007). I expected that roots would be able to grow into receptor site soil, but I predicted that root growth of both species in this investigation would be lower in receptor site soil than in donor site soil, especially within the barren areas, as I suspected plant available metals would be highest in this zone. Root mass density of *C. borealis* and *G. procumbens* grown in receptor site soil within the barren zone was found to be 25% and 19% smaller, respectively, when compared to the root growth of the same species grown in donor site soil, but the difference was not statistically significant (p= 0.129) (Figure 9, Table 6). Within the semi-barren zone there was no significant difference in root growth in donor and receptor site soil.

Findings from the stepwise regression analysis partially support my prediction that root growth would be reduced in plots closer to the smelter because variation in root mass density (mg_{root}/cm^3_{soil}) of *G. procumbens* was partially explained by plant available nickel (Table 7, Table 8). It is important to mention that plant available Ni was a retained variable from the principal component analysis and represented the variation of plant available Co and Cu in this investigation such that plots with high concentrations of plant available Ni also had high concentrations of plant available Co and Cu. The negative relationship between root growth in *G. procumbens* and plant available nickel in this model is supported by a study by Hutchinson & Whitby (1973) that found root elongation of vegetable species to be lower when plants were grown in a soil solution derived from soils collected close to the Sudbury smelters. Reduced root growth in the Hutchinson & Whitby (1973) study was caused by increased Cu and Ni uptake. Findings from my study suggest that root growth of this species might be lower in plots with elevated concentrations of plant available Ni. However, because there are no significant differences between root growth in donor and receptor site soil, I do not expect root growth to be limited to an extent that will be detrimental to establishment.

In contrast to *G. procumbens*, root growth of *C. borealis* could not be explained by variables related to smelter emissions. Root mass density of *C. borealis* was explained only by soil growing degree days; such that root growth of this species was greater in plots with higher soil temperatures (Table 7, Table 8). Roots of *G. procumbens* had a similar relationship with soil growing degree days; however, as mentioned previously, part of this variation was also explained by plant available nickel (Table 7, Table 8). The relationship between soil temperature and root growth in *C. borealis* is consistent with the literature. In general, root growth is known to be a function of soil temperature (Kaspar & Bland 1992) such that if moisture and nutrient status of the soils are adequate, root elongation will increase with increasing temperature until it reaches the optimal temperature for root growth (Pregitzer *et al.* 2000). I suspect that *C. borealis* roots did not respond to metal stress as strongly as *G. procumbens* because it has a deeper root system (Donohue *et al.* 2000; Mallik & Karim 2008) and was able to avoid heavy metal stress by producing roots in deeper soil layers that have lower concentrations of metals (SARA-Group 2001; Salemaa & Monni 2003). Past vegetation surveys have also found *C. borealis* to be capable of occupying smelter-affected lands within the region (Gordon & Gorham 1963). Differences in variables explaining root growth of these two species could also be caused by differences in their mycorrhizal association (Leyval *et al.* 1997; Pawlowska & Charvat 2004). *Clintonia* sp. has been shown to be associated with arbuscular mycorrhizal fungi whereas *G. procumbens* has been found to be associated with ericoid mycorrhiza (Malloch & Malloch 1981; DeBellis & Widden 2006); both of which are known to reduce the effects of metal stress on the plant host (Leyval *et al.* 1997). However, further studies would be required to test this hypothesis.

I had expected soil temperature to be related to percent canopy cover of the site; however, Pearson's product moment correlation did not find this relationship (Appendix B). It is difficult to determine what is driving the variation in soil temperature in my experiment because it is known to be influenced by air temperature, snow cover, soil moisture, soil type, soil density, latent heat exchange and solar radiation changes at and near the ground surface (Farouki 1981). The observed relationship seen in this study might also be caused by greater decomposition rates in sites with warmer soils. This relationship between temperature and decomposition is supported by a significant weak positive correlation between total inorganic nitrogen and soil growing degree days in my experiment ($r = 0.55$, $p<0.05$). Other studies have shown that warm sites had higher decomposition rates that led to increased nitrogen availability due to increased mineralisation at these sites, that led, in turn, to subsequent improved root growth (Clarkson *et al.* 1986; Bonan & van Cleve 1992). These findings suggest that warm sites could be advantageous for establishment of reintroduced *Clintonia* sp. and *Gautheria* sp. in the Sudbury region; however, the data in the current project do not provide conclusive evidence regarding factors leading to the observed variation in temperature. The growth-promoting effects of high soil temperatures would indicate an advantage of transplantations on south-facing slopes, but one has to be aware of the potential negative effects of drought on such sites (Warren II 2008). Studies looking at the influence of slope aspect on transplant survival in understory herbs found that the survival of *Hexastylis arifolia* was greatest on south facing slopes provided that they were shaded (Warren II 2010). In my investigation I observed that plots with less than 60% canopy cover appeared to be showing signs of drought stress. These signs included dead or yellowing leaves of individuals within the plots. Hence, I recommend future studies to determine factors driving this variation in soil temperature observed in this investigation as they would be helpful for placement of transplant material in the future.

I suspect that differences in root growth in barren and semi-barren zone could not be detected by the split-split plot analysis due to confounding effects of site recovery since the delimitation of zones in 1973. The time since restoration took place as well as the type of restoration that occurred varies from site to site in this investigation; for example, some plots were planted with red pine and others have received no restoration at all. Sites located within the barren zone described using air photos by Struik in 1973 have recovered due to re-greening activities and smelter emissions reductions that have occurred during this time (Sherman 2005; Environment Canada 2010 from Watson *et al.* 2012, McCall *et al.* 1995). I also suspect that as sites recover from metal pollution and low pH impacts, other site characteristics such as soil temperature become important in regulating root growth. This idea is supported by an unpublished study that took place in Coniston that found that once soil was detoxified using limestone application that phosphorus and nitrogen became the primary factor limiting plant growth (Farkouh 1978 as cited by Winterhalder 1996). I argue that the limits of the barren zone outlined in 1973 are out-dated and should be re-drawn to reflect the remediation that has taken place over the years.

5.2 Environmental variables as predictors of sexual reproduction in transplanted species

In general, variables related to both smelter emissions and other environmental site characteristics influenced the number of individuals that produced fruits and flowers within the plots, but these relationships were species dependent. The relationships between indicators of successful establishment for each species and the application of these findings to applied restoration will be discussed at the end of the discussion chapter.

Clintonia borealis

I found that the number of fruiting individuals of *C. borealis* was negatively correlated with the amount of sulphate within the receptor site soil and positively correlated with the amount of plant available nickel (Table 8). The relationship between the number of fruiting individuals and soil sulphates is similar to other studies. Cerdá (1984) who grew *Lycopersicon esculentum* Mill. Var hybrid 6C-204 in a nutrient solution containing SO_4^{2-} concentrations (ranging from 0 to 105 me/L) found that fruit yield in grams per plant decreased with increasing SO_4^{2-}, but the number of fruits per plant varied with concentration. It is important to note that the relationship between soil sulphate and establishment of species in this investigation is related to the role of sulphur emissions in increasing the availability of plant available metals in this region (Hutchinson & Whitby 1974; Greger 2004).

As stated earlier, plant available nickel was also found to have a positive effect on the number of fruiting *C. borealis* individuals in the plots. I had predicted that the number of fruiting individuals would be lower in plots with higher concentrations of plant available Ni as found in previous studies of *Hieracium piloselloides* and *Potentilla anserina* grown in soils with elevated concentrations of Cu and Ni (Saikkonen *et al.* 1998; Ryser & Sauder 2006). Plants are known to respond to stress by increasing investment into sexual reproduction as a way to disperse out of unfavourable growing conditions (Loehle 1987). *C. borealis* could be responding to metal stress by investing more into sexual reproduction. This explanation is supported by studies in

Monchegorsk, Russia that found individuals of *Vaccinium myrtillus*, *V. vitis-idaea,* and *Betula pubescens s*ubsp. *Czerepanovii* produced more seeds when growing in close proximity to a Cu-Ni smelter compared to those growing farther away (Zvereva & Kozlov 2004; Zvereva &Kozlov 2005). I had not expected sexual reproduction in *C. borealis* to respond so strongly to smelter emissions because neither SO_4^{2-} (p=0.56) or Ni (p= 0.48) explained variation in root growth in this species. The production of flower stalks and fruits in this species is very demanding in terms of carbon investment (Pitelka *et al.* 1985). I suspect that in plots with elevated concentrations of SO_4^{2-} and Ni the stress was too great and the plants could no longer afford to invest in sexual reproduction. I am unsure if this will actually affect spread of *C. borealis* in the future, because it primarily spreads through vegetative means (Angevine & Handel 1986; Dorken & Husband 1999). I would recommend that this species continue to be placed in sites within 5 km of the three current and historic smelters that have received limestone application because studies have identified this species growing within 5 km of the Falconbridge, Ontario smelter (Gordon & Gorham 1963). However, long-term cover and abundance surveys of *C. borealis* should be implemented to determine if stress at sites with elevated SO_4^{2-} and Ni concentrations will actually limit vegetative reproduction.

Maianthemum canadense

Variation in the number of flowering *M. canadense* individuals was best predicted by distance of the plot from the smelter and plant available phosphorus. The number of flowering individuals was greater in plots that were farther away and were lower in plots with more plant available phosphorus (Table 7, Table 8). I had predicted the positive relationship between the number of flowering individuals and distance from the smelter because other studies have found that in close proximity to Cu-Ni smelter colonization of understory plants is limited (Salemaa *et al.* 2001). Sudbury soils have the highest concentrations of metals in the top 5 cm of soils, and because *M. canadense* has shallow roots, I expected metals negatively to affect growth, particularly in sites close to the smelter (Flinn & Pringle 1983; SARA-Group 2001). Soil acidification is also known to promote the leaching of nutrients from upper soil horizons in smelter-affected soils (Kozlov & Zvereva 2007). A study of the influence of Cu-Ni smelter emissions on environmental site characteristics in South-Western Finland and North Western Russia found sites in close proximity to smelters had lower tree stand density, snow depth, percent water content in organic soils and exchangeable nutrients, respectively, in comparison to sites farther away. Sites closer to the two smelters in this study also had higher light availability and wind speeds (Zvereva & Kozlov 2001). In Sudbury, limited seedling establishment close to the smelter has also been explained by harsh microclimate caused by poor canopy cover (James & Courtin 1985). The ability of understory species naturally to colonize degraded sites close to the smelter in South-Western Finland was found to be limited because of elevated concentration of soil metals, nutrient imbalance and reduced water-holding capacity of sites close to the smelter (Salemaa *et al.* 2001). Findings from my study suggest it is less likely that *M. canadense* will be capable of spreading from plots close to the smelter using sexual reproduction. However, because this species can spread using vegetative reproduction (Sobey & Barkhouse1977; Silva *et al.* 1982) long-term studies will be needed to see the effects of distance from the smelter on vegetative reproduction.

I had not expected that plots with higher concentrations of plant available phosphorus would produce less flowers because generally plants become more productive with increased nutrient addition (Chapin III *et al.* 1986). This negative relationship has several possible explanations. It can be a coincidental result of decreased investment in sexual reproduction resulting from better growing conditions farther from the smelter, as seen in *Clintonia borealis*. Plant available phosphorus is higher farther from the smelter in sites that have not received limestone application. This is because plant available phosphorus in the soil is known to become less available as a result of limestone application (Schachtschabel *et al.* 1989). This negative relationship can be seen in my investigation between phosphorus and calcium ($r = -0.38$, $p<0.05$). Another possible explanation is that the reduction in the number of flowering *M. canadense* could be caused by increased competition in plots with more plant available phosphorus. When species with inherently low growth rates, such as understory herbs, are grown in nutrient-rich environments they can become competitively eliminated by fast growing species (Grime 2001; Chapin III *et al.* 1986). This was seen in a 10-year study in which tall grasses and herbaceous dicots began to dominate a site following nutrient addition leading to the subsequent reduction in *Linnaea borealis* and *Arctostaphylos uva-ursi* (Turkington *et al.* 2002). However, in my investigation the number of non-target weedy species occupying the plots was not explained by the plant available nutrients. It is unclear how these findings will affect the spread of *M. canadense* in the future as this species can spread both through sexual and vegetative means (Worthen & Stiles 1986; Hisatomo *et al.* 2008, Sobey & Barkhouse1977; Silva *et al.*1982).

Gaultheria procumbens

BpH and percent canopy cover were both significant predictors of variation in flowering frequency of *Gaultheria procumbens*. The frequency of flowering individuals within the plot was negatively related to BpH and positively related to canopy cover (Table 7, Table 8). As mentioned in the results chapter, BpH was retained as a predictor variable in place of pH because pH could not be transformed to attain assumptions of normality for regression analysis. The negative relationship between flowering frequency and BpH is supported by the literature, as this species is known to have an affinity for acidic soil (Carleton & Maycock 1981; Whitney & Foster 1988; Moola & Vasseur 2009). I had not expected the frequency of flowering to be lower in plots with higher canopy cover in my experiment because this species is frequently found in dry and open habitats (Curtis 1959; Moola & Vasseur 2004; Moola & Vasseur 2009). In *Vaccinium myrtilloides Mix*, another Ericaceous species typically found in open habitats, sexual reproductive output has also been found to be greater under partially shaded conditions (Moola & Mallik 1998). Moola & Vasseur (2009) also found *G. procumbens* growing in young regenerating forests (5% cover) to produce 83 more flowers/m^2 in comparison to an open site with no cover. Although canopy covers in my experiment were much greater (ranging 51% - 92%) than found in the Moola and Vasseur study, it is likely that some canopy cover will be favourable for sexual reproduction in *G. procumbens*. Based on my findings, the best sites to place *G. procumbens* are those with slightly acidic soils that have some canopy cover.

As minimum winter soil temperatures are known to be much lower (- 6.8 °C) in smelter-affected sites than less affected sites farther away (-0.2 °C) (Zvereva & Kozlov 2001), probably due to reduced plant cover, I had predicted that soil temperatures would be lower in sites closer to the smelter and that this would be detrimental to establishment. However, the lowest soil temperature during the winter months for most of my plots ranged between 0°C and -2 °C at a depth of 2 cm below the soil surface. Only 7 plots had temperatures that dipped below -5 °C, but only two of these plots remained at this temperature for more than two days in a row. I do not expect soil temperatures of the plots in my study to have dipped to low enough temperatures for a long enough period of time to cause damage to roots that would affect growth. An eight-year snow removal study found mean winter soil temperature of areas cleared of snow to be -5.5 °C at a depth of 10 cm below the soil surface and that soil temperature of the control ranged between -1.9 and -0.3 at this same depth. Root damage caused in the snow removal treatment caused 50% reduction of understory cover in comparison to the control (nearly 100% cover) (Kreyling *et al.* 2012). Thus, I do not expect winter soil temperatures to affect root growth of understory species in my study.

5.3 Diversity, richness, and species spread out of plot

Environmental site characteristics did not explain variation in species richness, Shannon-Wiener diversity index, evenness or the number of species spreading from the plot. I expected the pollution gradient caused by smelter emissions to limit species richness and Shannon-Wiener diversity index as seen in previous studies of smelter damaged lands (Freedman & Hutchinson 1980; McLaughlin 1985). However, this was not the case. Although not tested statistically because only six donor sites were observed in this investigation, it does appear that species richness was higher in receptor plots than donor sites. Average species richness for receptor plots was 23.5 and 17.2 for donor plots. The greater average species richness in receptor sites was probably caused by the attempt of the reclamation workers installing the mats to reintroduce as many species that were ecologically appropriate to each site. However, it does not appear that this had an influence on Shannon-Wiener Diversity Index. The Shannon-Wiener Diversity was 2.65 for receptor plots and 2.4 in donor site plots. Variation in species evenness within the plot was weakly predicted by distance of the plot from the smelter ($r^2 = 0.12$, $p = 0.065$), but the range in species evenness within transplant plots (0.67 to 0.96) does not appear to be different from that of the donor sites (0.72 to 0.93). As vegetative reproduction has been found to be limited in soils exposed to elevated metal associated with smelter emissions (Saikkonen *et al.* 1998) I had expected that the number of individuals that spread from the plot would be related negatively to smelter related variables. However, this was not found in my experiment. The number of individuals that spread from receptor plots ranged between 0 and 26 individuals, but on average 3.5 individuals spread from these plots. It is encouraging that variation in species richness and diversity was not seen in the first summer following transplantation. Because of damage to roots caused by the process of transplantation, I had expected that newly transplanted species would be more susceptible to stresses within transplant plots during their first year of growth. For example, Ashmun & Pitelka (1985) found mortality rates were the highest during the winter following the first year of growth (12% to 20%) for *Clintonia borealis* transplanted into experimental gardens. In my experiment, conditions within plots were likely not stressful enough to cause massive die-offs in newly introduced species during their first growing season. However, because this is a short-term study it is likely too early to see changes in species composition and population growth of transplanted vegetation caused by differences in plot microenvironment.

The number of non-target species within the plots also could not be explained by environmental characteristics. Weedy, non-native vascular plant species are known to occupy restored sites within the Sudbury region, thus I was concerned that metal-resistant fast-growing "weedy" species present in the Sudbury area would begin to colonize nutrient rich mats and out-compete slow-growing understory species (Rayfield *et al.* 2005; Chapin III *et al.* 1986). Twenty-one receptor plots had non-target species within them but on average only 1.2 non-target species were found within the plots. The majority of these species were either *Hieracium* sp. or *Rumex acetosella*. Only one plot in the donor site had one non-target individual, *Melampyrum pratense,* that although native to Europe, is a slow growing species that typically occupies low nutrient forests (Smith 1963). Long-term monitoring should be implemented to ensure that fast growing species do not out compete transplanted species because 70% of my plots had at least one non-target individual in them.

I caution that this was a short-term study and it is too early to see changes in species composition and population growth of transplanted vegetation caused by differences in plot microenvironment. Looking at root growth and investment into sexual reproduction provided valuable insight into growth response to environmental characteristics in the short-term. However, long-term monitoring of the further development of these plots will be needed to truly gauge the success of this project. Drayton & Primack (2012) stress the importance of using long-term studies to monitor population establishment because it takes many years, particularly for perennials, to reach reproductive maturity. This approach is supported by restoration projects using transplanted native species by Fattorini (2001) and Peterson & Philipp (2001) who found greater changes in species survival and evidence of new seedling establishment after ten years in comparison to three years following transplantation. Furthermore, basing establishment on high rates of initial growth is cautioned in rhizomatous herbs, because initial production of new ramets could be supported by carbohydrate reserves. Long-term establishment of rhizomatous plants is only sustainable if species are unable to adapt to a new environment (Nelson *et al.* 2007). Therefore, I suggest that long-term monitoring be continued so that true measures of successful reintroduction, the establishment of viable populations, can be evaluated.

5.4 Application of findings to applied restoration

In the following section I will discuss application of my findings to applied restoration as well as recommendations for future transplant projects within the Sudbury region. The first two points relate to the placement of understory species within sites in the future. My last three points deal with strategies to encourage the colonization of understory species into smelter disturbed sites, long-term monitoring strategies and a recommendation to up-date Struiks' barren and semi-barren map created in 1973.

1) Using mats of salvaged vegetation and associated soil placed in large 16 m^2 plots was found to be a feasible method of reintroducing understory species into smelter-affected forests of Sudbury. This is because, in general, plants appear to have established with little signs of stress. However, some environmental site characteristics were found to influence successful establishment; namely, smelter related variables and soil temperature. Based on my findings most understory species will perform best in warm sites (south-facing slopes) in smelter affected forests provided they have an adequate canopy cover. I would suggest that species be placed under at least partial canopy cover because plants in plots with less than 60% cover appeared to show signs of drought stress. Species typical of open disturbed sites, such as *Gaultheria procumbens*, are expected also to perform better in partial-shade conditions.

2) Restoration efforts have confounded the difference between the barren and semi-barren zones, which were described in the early 1970s. The transplantation of understory species should be continued in areas with appropriate canopy cover within the "barren zone".

Root growth in both observed species may have been lower in the barren zone, but these differences were not significant. Moreover, smelter related variables were not found to explain variation in species richness or diversity within the plots. However, the sensitivity of *G. procumbens* root growth to higher concentrations of plant available nickel indicates that some species might be negatively affected by metal stress. Hence, species with known metal tolerance should be favoured for transplantation in sites with elevated metals or soil sulphates. Information regarding potential for metal tolerance in understory is available from previous vegetation surveys within the metal stressed environments within this region. I would recommend using an indicator species approach when introducing understory species in the future. The idea that plants can be used as an indicator of environmental conditions within sites is not a new concept (Klinka *et al.* 1989, Ringius & Sims 1997). This approach is based on the fact that each plant species is adapted to a range of particular environmental conditions and its distribution is restricted to sites that lie within this range. Thus, knowing the ecology of plant species that reside in particular sites make it possible to infer a sites qualities (Klinka *et al.* 1989, Ringius & Sims 1997). Table 9 provides a list of candidate understory species based on scientific literature and vegetation surveys performed throughout the region that I would recommend planting within metal-stressed areas with appropriate canopy cover but that currently contain hardly any shade tolerant vegetation. I would also advise against introducing species that are uncommon to the donor site or species that are known to be intolerant to metal stress in restored forests that do not contain any shade tolerant vegetation already. Table 10 provides a list of potentially metal-sensitive species or plants that were uncommon in donors site that should be placed in areas that are less metal-stressed and that already contain indicator shade tolerant species and thus indicate favourable growing conditions. Examples of such species include: *Cornus canadensis*, *Maianthemum canadense*, or *Clintonia borealis* monocultures.

Table 9: Understory species capable of tolerating metal stressed sites.

Metal resistant species		**Source**
Herbs		
Aralia nudicaulis	wild sarsaparilla	Gordon & Gorham 1963
Aster macrophyllus	large leaf aster	Gordon & Gorham 1963
Clintonia borealis	bluebead lily	Gordon & Gorham 1963
Cornus canadensis	Canadian bunchberry	Gordon & Gorham 1963
Epigaea repens	trailing arbutus	Santala & Popp 2010 *unpublished data*
Fragaria virginiana	wild strawberry	Santala & Popp 2010 *unpublished data*
Maianthemum canadense	Canada mayflower	Gordon & Gorham 1963
Trientalis borealis	northern starflower	Gordon & Gorham 1963
Pyrola sp.	shinleaf	Santala & Popp 2010 *unpublished data*
Shrubs		
Aralia hispida	bristly sarsaparilla	Santala & Popp 2010 *unpublished data*
Amelanchier sanguinea	serviceberry	Gordon & Gorham 1963
Corylus cornuta	beaked hazel	Gordon & Gorham 1963
Diervilla lonicera	bush-honeysuckle	Gordon & Gorham 1963
Gaultheria procumbens	wintergreen	Santala & Popp 2010 *unpublished data*
Ilex verticillata	common winterberry	Santala & Popp 2010 *unpublished data*
Grass and Sedges		
Deschampsia flexuosa	wavy hair grass	Gordon & Gorham 1963
Oryzopsis asperifolia	white-grain mountain-rice grass	Gordon & Gorham 1963
Lichen and Mosses		
Cladonia sp.		SARA-Group 2009
Cladina sp.		SARA-Group 2009

Table 10: Understory species known to be sensitive to metal stress or are uncommon to the donor site.

Metal sensitive		**Source**
Herbs		
Agrimonia gryposepala	hairy agrimony	Santala & Popp 2010 *unpublished data*
Cypripedium acaule	pink lady's slipper	Santala & Popp 2010 *unpublished data*
Coptis trifolia	threeleaf goldthread	Santala & Popp 2010 *unpublished data*
Fragaria vesca	woodland strawberry	Santala & Popp 2010 *unpublished data*
Galium triflorum	fragrant bedstraw	Santala & Popp 2010 *unpublished data*
Geranium bicknellii	Bicknell's cranesbill	Santala & Popp 2010 *unpublished data*
Hepatica americana	round-leaved hepatica	Santala & Popp 2010 *unpublished data*
Lathyrus ochroleucus	pale vetchling	Santala & Popp 2010 *unpublished data*
Maianthemum racemosum	false Solomon's sea	Rayfield *et al.* 2005
Melampyrum lineare	narrowleaf cowwheat	Santala & Popp 2010 *unpublished data*
Polygala paucifolia	gaywings	Santala & Popp 2010 *unpublished data*
Waldsteinia fragarioides	barren strawberry	Rayfield *et al.* 2005
Streptopus lanceolatus var. roseus	rose-twisted stalk	Santala & Popp 2010 *unpublished data*
Viola sp.	forest violets	Santala & Popp 2010 *unpublished data*
Shrub		
Aronia melanocarpa	black chokeberry	Santala & Popp 2010 *unpublished data*
Chimaphila umbellata	prince's pine	Santala & Popp 2010 *unpublished data*
Lonicera canadensis	American fly honeysuckle	Rayfield *et al.* 2005
Linnaea borealis	twinflower	Santala & Popp 2010 *unpublished data*
Mitchella repens	partridgeberry	Santala & Popp 2010 *unpublished data*
Rosa acicularis	prickly rose	Santala & Popp 2010 *unpublished data*
Viburnum nudum var. cassinoides	wild rasin	Santala & Popp 2010 *unpublished data*
Grasses, Sedges and Rushes		
Carex sp.		SARA-Group 2009
Brachyelytrum erectum	bearded shorthusk	SARA-Group 2009
Luzula acuminata	hairy woodrush	Santala & Popp 2010 *unpublished data*

Table 10: Continued: Understory species known to be sensitive to metal stress or are uncommon to the donor site.

Metal sensitive		**Source**
Lichen, Mosses and Bryophytes		
Brachythecium sp.	brachythecium moss	Santala & Popp 2010 *unpublished data*
Cladonia multiformis		SARA-Group 2009
Dicranum sp.	dicranum moss	SARA-Group 2009
Hypnum sp.		SARA-Group 2009
Leucobryum glaucum	leucobryum moss	Santala & Popp 2010 *unpublished data*
Platygyrum repens		Santala & Popp 2010 *unpublished data*
Pleurozium schreberi	Schreber's big red stem moss	Santala & Popp 2010 *unpublished data*
Sphagnum sp.	sphagnum	SARA-Group 2009
Ferns and Allies		
Dryopteris carthusiana	spinulose woodfern	Santala & Popp 2010 *unpublished data*
Huperzia lucidula	shining clubmoss	Santala & Popp 2010 *unpublished data*
Lycopodium annotinum	clubmoss	Santala & Popp 2010 *unpublished data*
Lycopodium dendroideum	tree groundpine	SARA-Group 2009
Onoclea sensibilis	sensitive fern	Santala & Popp 2010 *unpublished data*
Osmundastrum cinnamomea	cinnamon fern	Santala & Popp 2010 *unpublished data*
Osmunda claytoniana	interrupted fern	Santala & Popp 2010 *unpublished data*

3) Because some measures of successful establishment were related negatively to smelter emissions, I would recommend engaging in restoration practices that would improve microclimate in smelter-disturbed forests close to the smelter. Three factors which limit understory establishment in Cu-Ni affected forests is elevated soil metal content, poor nutrient availability and reduced water-holding capacity (Salemaa *et al.* 2001, Kozlov & Zvereva 2007). I recommend the planting of native deciduous trees and shrubs, as it would help alleviate some of these stresses by building organic matter content (Sherman 2005). The Sudbury Ecological Risk Assessment study pointed out that low organic

matter content is a contributing factor to the poor recovery of plant species richness within this region. However, they noted that it was impossible to separate completely its influence on the plant community with other correlated variables such as soil erosion, low pH, and soil fertility (SARA-Group 2009). Leaf fall would increase organic matter content which could subsequently help reduce the solubility of plant available metals and other chemicals of concern, maintain soil fertility, and improve water holding capacity (Haghiri 1973; Khaleel *et al.* 1981; Raviv 1986; Senesi & Sposito 1989; Pulford & Watson 2003).

4) This study provided insight into short-term responses of vegetation to microclimatic conditions one year following introduction; however, long-term studies will need to be continued so that true measures of successful reintroduction, namely, the establishment of viable populations, can be evaluated. The duration of monitoring programs varies from study to study. Drayton & Primack (2012) reported that 2 years following introduction, 7 of 8 introduced individuals were still present in plots, but 15 years later only 2 of the originally introduced species had survived. Fattorini (2001) found that in plots installed in 1995 and 96, between 86% and 95% of species survived the first 2 to 3 years. He found for plots installed in 1985 and 86 that all transplanted species were still present after 12 to 13 years. Peterson & Philipp (2001) found that *Mercurialis perennis* L. and *Corydalis bulbosa* L. (DC.) spread approximately 50 cm after the first 2 years of growth, and 125 cm 10 years later. I would recommend that a portion of the sites be monitored yearly for ruderal species cover for at least the first 5 years to ensure fast growing species do not take over the plots. If ruderal cover exceeds 30% of the plot, I would recommend weeding the plots or making adjustments to planting technique based on preliminary

findings. After the first 5 years I recommend that plots be evaluated once every 5 or 10 years to track changes within the plot. Reviews of transplant projects by Godefroid *et al.* (2011) found the duration of long-term programs range between 10 years to several decades depending on the project, but that the length of the program should reflect the generation time of the species involved. Although understory plants spread primarily through vegetative reproduction, seeds are required to establish a new population. It can take up to 10 years, depending on the species, to reach an age at which they can reproduce sexually (Bierzychudek 1982). Therefore, I recommend the duration of monitoring exceed 20 years to acquire conclusive results. Long-term monitoring should include, but is not limited to, measures of cover and composition of target species within the plot and spread of target species out of the plot. I also recommend that site characteristics be re-evaluated to determine if changes in the reclaimed forests have an influence on transplant establishment (increased cover, insect defoliation of canopy or climate change). In doing this, if species do well or poorly in relation to the sites in which they are put, science-based adjustments to future transplants can be made to build on successes and to avoid failures. It is important to note that in addition to a corner post, nails have been placed at the corner of ever plot such that, in the event a corner post being lost, the original extent of the plot can be determined with a metal detector.

5) I recommend a revised map be created that describes the current extent of damage and recovery within the Sudbury area. Struiks' map created in 1973 is very outdated and no-longer accurately describes limits of the barren and semi-barren zones. My argument is not new. Sinclair (1996) who characterized plant communities in the Sudbury region argued that the term "barren" should no longer be used to describe "barren sites"

described by Amiro & Courtin (1981) as these areas could support a diverse community of metal tolerant grasses and non-vascular plants. My findings support hers, as all my sites within the “barren” zone were located under the canopy of an anthropogenic created forest and hence are inconsistent with the definition of barren zone. Reviews by Kozlov & Zvereva (2007) add to this argument that industrial barrens described as having even 5% or 10% cover may be misleading, particularly in areas with naturally low vegetation cover. I suggest we adopt a new definition for industrial barrens described by Kozlov & Zvereva (2007) that is more applicable locally and internationally. They classify industrial barrens as bleak open landscapes that have developed around point sources of industrial pollution due to deposition of airborne pollution. These areas are comprised of small patches of vegetation (less than 10% cover in comparison to reference sites) surrounded by bare land comprised of alluvial horizon or even rock exposed to intensive soil erosion (relict soil less than 20%). Restoration ecologist and researchers would benefit from having a publically accessible up-to-date map that accurately delimits the extent of damage and recovery within the Sudbury area.

5.5 Conclusions

In this study I was able to demonstrate that environmental variables related to smelter emissions or other natural environmental variables predicted species establishment, and that these relationships were species specific. The introduction of understory species should be continued even within 5 km of current and historic smelters because smelter related variables were not found to affect species diversity, richness, spread of species and because the location of plots within each smelter-disturbance zone did not strongly influence root growth. Even in the most metal-stressed sites within this study it appears that at least some of the understory species were capable of establishing. Based on my findings understory species will perform best in warm sites with adequate canopy cover. However, I suggest that only species known to tolerate metal stress be placed in smelter-stressed sites because some smelter-related variables were found to negatively affect root growth of *G. procumbens*, and number of flowering individuals of *C. borealis* and *M. canadense*. Information gathered through this study is useful for site selection when transplanting understory species in the future Canadian regreening projects as species in this study can be found in both boreal and temperate forests. However, it is important to note that this study provides insight only into short-term responses of vegetation to microclimate conditions; and that long-term studies will need to be continued so the establishment of viable populations, the true measure of successful establishment of understory species, can be evaluated.

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Appendices

Appendix A: Date and climate conditions at time of transplantation at the Sudbury Airport (46 37"N, 80 48"W, 347.5 m above sea level) in 2010 (Environment Canada, 2013).

Site	Plot	Date Planted	Average Temperature (°C)	Precipitation (mm)
1	2	2010-05-27	22.1	0.2
1	3	2010-05-27	22.1	0.2
3	1	2010-06-16	12.1	14.6
3	2	2010-06-16	12.1	14.6
3	5	2010-06-16	12.1	14.6
5	2	2010-06-08	12.3	0
5	4	2010-06-08	12.3	0
5	5	2010-06-08	12.3	0
6	2	2010-06-14	17	1.4
6	4	2010-06-17	16.9	0
6	5	2010-06-17	16.9	0
8	3	2010-06-20	18	0
8	4	2010-06-20	18	0
8	5	2010-06-22	15.3	10.6
10	1	2010-06-23	19.2	3.4
14	3	2010-07-08	23.8	0.8
15	2	2010-07-21	20.4	1.4
15	5	2010-07-21	20.4	1.4
22	1	2010-07-28	20.8	4.8
22	3	2010-07-28	20.8	4.8
22	5	2010-07-24	20.1	0
25	5	2010-08-16	17.5	13.8
26	1	2010-08-16	17.5	13.8
26	3	2010-08-16	17.5	13.8
34	2	2010-09-02	20.5	trace
34	4	2010-09-02	20.5	trace
34	5	2010-09-02	20.5	trace
36	3	2010-09-29	9.5	trace
36	4	2010-09-29	9.5	trace
36	5	2010-09-29	9.5	trace

Appendix B: Pearson Product-moment correlations between environmental predictors; Abbreviations are as follows: Log preceding variables indicates it was $\log_{10}$ transformed, As is arsenic, Se is selenium, Pb is lead, Co is cobalt, Ni, sGDD is soil growing degree days, BpH is buffering pH, C.E.C is cation exchange capacity, OMG is organic matter content, P is phosphorus, K is potassium, Mg is Magnesium, Ca is calcium, SO_4^{2} is soil sulphate, NO^{3-} is nitrate, NH^{3-} is ammonium, distance is distance of plot to closest smelter (km). Significant correlation are **bolded** (p<0.05).

	logSe	logPb	logCo	logNi	logCu	Canopy Cover	sGDD	pH	BpH	C.E.C	logOMC	Log P	log K	log Mg	Log Ca	$\log SO_4^{2}$	$\log NO^{3-}$	$\log NH^{3-}$	log Distance
logAs	**0.67**	**0.82**	0.25	0.02	0.20	0.03	0.01	**-0.46**	**-0.45**	**0.44**	**0.61**	0.18	0.10	0.00	-0.09	-0.27	**0.44**	0.25	**0.48**
logSe		**0.49**	0.26	0.25	**0.68**	-0.01	0.11	**-0.40**	**-0.43**	**0.41**	**0.51**	0.16	0.04	-0.05	-0.14	-0.08	0.15	0.28	0.14
logPb			**0.39**	0.21	0.12	0.18	0.03	-0.26	-0.26	0.28	**0.62**	0.05	0.14	-0.10	0.07	-0.26	**0.53**	**0.37**	**0.62**
logCo				**0.86**	0.35	**0.49**	-0.18	0.28	0.18	-0.08	0.24	-0.32	-0.19	0.30	0.15	0.16	0.06	-0.15	0.07
logNi					**0.47**	0.31	-0.15	**0.41**	0.25	-0.10	0.21	**-0.40**	-0.26	**0.42**	0.23	0.25	-0.04	-0.16	0.09
logCu						-0.03	0.05	-0.17	-0.24	0.22	0.21	0.09	0.02	-0.13	-0.02	0.00	0.00	0.17	-0.10
Canopy Cover							0.20	0.02	-0.10	0.20	**0.39**	**-0.39**	0.16	-0.02	-0.11	0.09	0.01	0.22	-0.18
sGDG								**-0.44**	**-0.47**	0.34	0.26	0.25	0.20	0.00	**-0.40**	-0.29	0.31	**0.57**	-0.05
pH									**0.95**	**-0.81**	**-0.49**	**-0.53**	-0.35	**0.40**	**0.55**	0.25	-0.15	**-0.58**	-0.16
BpH										**-0.93**	**-0.65**	**-0.42**	-0.36	0.31	**0.48**	0.21	-0.20	**-0.60**	-0.13
C.E.C											**0.77**	0.23	0.31	-0.18	-0.29	-0.16	0.22	**0.54**	0.13
logOMC												0.00	0.28	-0.10	-0.17	-0.32	**0.43**	**0.53**	**0.46**
logP													**0.52**	-0.25	**-0.38**	**-0.57**	0.30	**0.49**	0.16
logK														**-.386**	-0.29	**-0.41**	0.24	**0.60**	0.12
logMg															0.24	0.31	-0.09	**-0.39**	-0.06
logCa																0.14	0.18	-0.31	-0.16
$\log SO_4^{2}$																	**-0.55**	**-0.49**	**-0.37**
$\log NO^{3-}$																		**0.50**	0.36
$\log NH^{3-}$																			0.24

Appendix C: Linear regression analysis showing environmental independent variables ability to significantly predict variation in spread of species out of the plots and number of non-target species in the plot.

Table C1: Simple linear regression analysis showing environmental independent variables ability to significantly predict variation in spread of species out of the plots.

Variable	r^2	F	p
Log Nickel µg/g	0.02	0.56	0.460
Log Arsenic µg/g	0.00	0.002	0.963
BpH	0.01	0.26	0.616
Log Total Nitrogen ppm	0.01	0.16	0.695
Log Phosphorus ppm	0.02	0.47	0.500
Log Total Sulphates ppm	0.04	1.24	0.276
Log Organic Matter %	0.04	1.17	0.288
Soil Growing Degree Days	0.01	0.17	0.681
% Canopy Cover	0.000	0.00	0.985
C.E.C. [1]	0.04	1.13	0.296
LogPlot Distance from Nearest Smelter km	0.005	0.13	0.722

[1] Cation Exchange Capacity

Table C2: Simple linear regression analysis showing environmental independent variables ability to significantly predict variation the number of non-target species within plots.

Variable	r^2	F	p-value
Log Phosphorus ppm	0.92	2.82	0.104
Log Arsenic µg/g	0.03	0.91	0.349
Soil Growing Degree Days	0.09	2.6	0.119
Log Total Sulphates ppm	0.07	2.04	0.164
Log Organic Matter %	0.05	1.38	0.25
C.E.C	0.02	0.64	0.43
% Canopy Cover	0.029	0.807	0.377
Log Total Nitrogen ppm	0.001	0.33	0.857
Log Plot Distance from Nearest Smelter km	0.007	0.21	0.651
log Nickel µg/g	0.000	0.003	0.955
BpH	0.002	0.05	0.832

1 Cation Exchange Capacity

Appendix D: Linear regression analysis showing environmental independent variables ability to significantly predict variation in richness, Shannon-Wiener Diversity Index, evenness of transplant plots.

Table D1: Simple linear regression analysis showing environmental independent variables ability to significantly predict variation in species richness within transplant plots.

Variable	r^2	F	p-value
Log Total Sulphates ppm	0.012	0.336	0.567
Log Organic Matter %	0.001	0.028	0.869
Soil Growing Degree Days	0.00	0.008	0.931
Log Total Nitrogen ppm	0.038	1.103	0.303
Log Plot Distance from Nearest Smelter km	0.049	1.446	0.239
Log Plant available Arsenic µg/g	0.000	0.011	0.918
C.E.C[1]	0.016	0.459	0.504
Log Plant Available Nickel µg/g	0.084	2.556	0.121
BpH	0.017	0.479	0.494
Log Phosphorus ppm	0.017	0.489	0.49
% Canopy Cover	0.000	0.000	0.988

1 Cation Exchange Capacity

Table D2: Simple linear regression analysis showing environmental independent variables ability to significantly predict variation in Shannon-Wiener Diversity Index.

Variable	r^2	F	p-value
Log Plot Distance from Nearest Smelter km	0.012	0.342	0.563
Log Phosphorus ppm	0.038	1.119	0.299
Log Organic Matter %	0.018	0.502	0.484
Log Total Nitrogen ppm	0.062	1.962	0.172
Soil Growing Degree Days	0.000	0.01	0.919
Log Plant Available Arsenic µg/g	0.023	0.657	0.424
% Canopy Cover	0.004	0.103	0.750
BpH	0.004	0.114	0.738
Log Plant Available Nickel µg/g	0.06	1.774	0.194
C.E.C [1]	0.003	0.072	0.791
Log Total Sulphates ppm	0.01	0.283	0.599

[1] Cation Exchange Capacity

Table D3: Simple linear regression analysis showing environmental independent variables ability to significantly predict variation in species evenness within transplant plots.

Variable	r^2	F	p-value
Log Plot Distance from Nearest Smelter km	**0.12**	**3.70**	**0.065**
Log Total Nitrogen ppm	0.05	1.53	0.227
Log Organic Matter %	0.04	1.05	0.314
% Canopy Cover	0.07	0.18	0.675
Log Phosphorus ppm	0.02	0.59	0.450
Log Plant Available Nickel µg/g	0.021	0.607	0.443
Log Total Sulphates ppm	0.01	0.26	0.617
Soil Growing Degree Days	0.002	0.04	0.837
Log Plant Available Arsenic µg/g	0.05	1.35	0.254
BpH	0.000	0.00	0.986
C.E.C. [1]	0.000	0.00	0.993

[1] Cation Exchange Capacity

Appendix E: Descriptive Statistics of Variables Related to Transplant success

Table E1: Descriptive statistics related to transplant success within receptor plots

Variable	n	Range	Minimum	Maximum	Mean	Standard Error	Standard Deviation
Reproduction							
Fruiting *M. canadense*	30	14.00	0.00	14.00	4.23	0.79	4.31
Fruiting *C. borealis*	30	6.00	0.00	6.00	1.70	0.34	1.86
Flowering *G. procumbens*	30	9.00	0.00	9.00	4.23	0.45	2.47
Root Growth							
C. borealis Root Mass Density (mg_{root}/cm^3_{soil})	28	1.21	0.16	1.37	0.63	0.07	0.36
G. procumbens Root Mass Density (mg_{root}/cm^3_{soil})	30	1.43	0.06	1.49	0.68	0.07	0.36
Spread From Plot							
Number of Individuals	30	26.00	0.00	26.00	3.50	1.19	6.52
Number of Species	30	6.00	0.00	6.00	1.40	0.31	1.67
Species Composition							
Shannon - Wiener Index	30	1.33	1.76	3.09	2.65	0.06	0.33
Richness	30	20.00	14.00	34.00	23.53	0.90	4.92
Evenness	30	0.29	0.67	0.96	0.84	0.01	0.07

Table E2: Descriptive statistics related to transplant success within donor plots

Variable	n	Range	Minimum	Maximum	Mean	Standard Error	Standard Deviation
Reproduction							
Fruiting *M. canadense*	6	28.00	0.00	28.00	7.50	4.36	10.69
Fruiting *C. borealis*	6	4.00	0.00	4.00	1.33	0.61	1.51
Flowering *G. procumbens*	6	3.00	4.00	7.00	5.17	0.40	0.98
Root Growth							
C. borealis Root Mass Density (mg_{root}/cm^3_{soil})	6	2.28	0.31	2.59	1.12	0.32	0.78
G. procumbens Root Mass Density (mg_{root}/cm^3_{soil})	5	1.43	0.35	1.78	0.93	0.24	0.53
Spread From Plot							
Number of Individuals	-	-	-	-	-	-	-
Number of Species	-	-	-	-	-	-	-
Species Composition							
Shannon - Wiener Index	6	0.94	1.84	2.78	2.40	0.13	0.31
Richness	6	12.00	13.00	25.00	17.17	1.80	4.40
Evenness	6	0.21	0.72	0.93	0.85	0.03	0.07

Appendix F: Descriptive Statistics of Environmental Characteristics

Table F1: Descriptive statistics related to environmental variables within receptor plots

Variable	n	Range	Minimum	Maximum	Mean	Standard Error	Standard Deviation
Smelter Emissions							
Plot Distance from Nearest Smelter km	30	6.83	1.86	8.69	4.90	0.37	2.05
Total Arsenic µg/g	30	128.04	7.96	136.00	45.39	5.73	31.37
Plant Available Arsenic µg/g	30	1.04	0.04	1.08	0.31	0.05	0.29
Total Nickel µg/g	30	461.90	70.10	532.00	237.60	18.48	101.19
Plant Available Nickel µg/g	30	15.81	1.49	17.30	5.57	0.72	3.96
Total Sulphates ppm	30	70.90	10.70	81.60	24.53	2.75	15.07
Soil Chemistry							
BpH	30	1.50	4.90	6.40	5.48	0.09	0.47
Cation Exchange MEQ/100g	30	15.60	12.70	28.30	21.48	0.81	4.43
Organic Matter %	30	30.90	4.20	35.10	9.70	1.38	7.59
Phosphorus ppm	30	78.00	5.00	83.00	26.30	3.35	18.33
Total Nitrogen	30	62.80	2.10	64.90	18.48	2.77	15.16
Microclimate							
Soil degree days	29	327.50	396.50	724.00	553.15	17.41	93.78
% Canopy Cover	30	40.56	51.38	91.94	80.38	1.69	9.26

Table F2. Descriptive statistics related to environmental variables within donor plots

Variable	n	Range	Minimum	Maximum	Mean	Standard Error	Standard Deviation
Smelter Emissions							
Plot Distance from Nearest Smelter km	6	0.24	37.48	37.72	37.61	0.04	0.10
Total Arsenic µg/g	6	7.00	5.20	12.20	8.17	0.96	2.36
Plant Available Arsenic µg/g	6	0.23	0.18	0.42	0.27	0.03	0.08
Total Nickel µg/g	6	82.60	30.40	113.00	74.17	11.20	27.43
Plant Available Nickel µg/g	6	1.03	0.34	1.37	0.80	0.17	0.41
Total Sulphates ppm	6	3.30	6.70	10.00	8.62	0.48	1.17
Soil Chemistry							
BpH	6	0.50	4.60	5.10	4.82	0.07	0.17
Cation Exchange MEQ/100g	6	3.80	26.80	30.60	28.73	0.58	1.42
Organic Matter %	6	19.30	24.30	43.60	34.75	3.17	7.76
Phosphorus ppm	6	8.00	8.00	16.00	12.33	1.05	2.58
Total Nitrogen ppm	6	74.80	31.50	106.30	66.22	14.22	34.84
Microclimate							
Soil degree days	5	95.00	427.75	522.75	457.20	17.91	40.04
% Canopy Cover	6	11.96	79.20	91.16	83.66	1.94	4.76

Appendix G: Total and plant available chemicals of concern within barren, semi-barren and donor sites

Table G1: Descriptive statistics of total and plant available elements (µg/g) in soils surrounding plots located within two impact zones barren, semi-barren or the undisturbed donor sites.

Variable	n	Range	Minimum	Maximum	Mean	Standard Error	Standard Deviation
Barren[1]							
Total Arsenic	15	96.04	7.96	104.00	46.34	6.62	25.64
Plant Available Arsenic	15	1.04	0.04	1.08	0.26	0.07	0.29
Total Nickel	15	433.70	98.30	532.00	259.88	28.74	111.30
Plant Available Nickel	15	15.54	1.76	17.30	6.02	1.06	4.13
Semi-Barren[2]							
Total Arsenic	15	120	15.70	136.00	44.43	9.59	37.13
Plant Available Arsenic	15	0.80	0.13	0.93	0.36	0.07	0.28
Total Nickel	15	331.00	70.10	402.00	215.31	22.74	88.08
Plant Available Nickel	15	11.81	1.49	13.30	5.13	1.00	3.88
Donor [3]							
Total Arsenic	6	7.00	5.20	12.20	8.17	0.96	2.36
Plant Available Arsenic	6	0.23	0.18	0.42	0.27	0.03	0.08
Total Nickel	6	82.60	30.40	113.00	74.17	11.20	27.43
Plant Available Nickel	6	1.03	0.34	1.37	0.80	0.17	0.41

[1] Barren zones (Winterhalder 1995, based on Struik 1973) which was described in 1978 by Amiro and Courtin (1981) as principally being devoid of trees and sparse of vegetation (Figure 3.4)

[2] Semi-barren zone (Winterhalder 1995, based on Struik 1973) and was described in 1978 by Amiro and Courtin (1981) as being a transition zone between the barrens and the natural plant community

[3] Donor sites include six control sites approximately 50 km South of Sudbury that were located adjacent to areas where vegetation for transplant project was salvaged.